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# CO-ORDINATED TRIALS WITH PHENOTHIAZINE AGAINST NEMATODES IN LAMBS

**A Report prepared at the instance of the Agricultural Research Council of the United Kingdom**

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## 1.—Historical Review

Phenothiazine, or thiodiphenylamine, has been in use as an insecticide since 1934. When finely milled it is a soft bulky powder, usually of a greyish- or even bluish-green colour though the pure substance is said to be lemon-yellow. It is wettable with difficulty and only slightly soluble in water, though readily soluble in a large number of organic solvents, the solutions giving a surprising range of colours. The solid melts at or slightly above 181° C.

In 1938, following up the insecticidal work, Knipling (22)* found that small doses of phenothiazine given internally to cattle prevented the development of hornfly larvæ in their fæces. Published accounts of its use strictly as an anthelmintic have become very numerous since the first report, also in 1938, by Harwood *et al.* (18), who claimed it efficacious against *Ascaris* and *Oesophagostomum* in pigs. The rapidly increasing interest in its anthelmintic properties is reflected in the number of entries (shown in parentheses below) in the subject indexes of Helminthological Abstracts covering the literature of successive years; 1938 (1), 1939 (12), 1940 (39), 1941 (51); and the spate continues.

It is impracticable to summarize all these papers here, and unnecessary in view of the fact that there have recently been published a bibliography by the Imperial Bureau of Agricultural Parasitology (21) and a review by the Veterinary Bulletin (3). Suffice it to point out that, of the 12 papers published in 1939, 10 were concerned with phenothiazine in sheep; good results were reported from Canada (Swales, 42), Australia (Gordon, 6, 7, 8, Gordon & Whitten, 10, Roberts, 33, 34), and the U.S.A. (Habermann & Harwood, 13, and Harwood *et al.*, 16, 17). The 1940 papers show the use of the drug extended, with favourable results, to cattle, horses, fowls and man. There are reports on the excretion by the host of phenothiazine or its derivatives, and at least one tentative explanation of its mode of action, by inhibiting hæmin catalysts (Collier, 2). Most of these earlier papers agree in finding the drug safe and effective against a large number of worms. And, although in some reports the actual evidence is scanty, or not critically sifted, or even not disclosed, there remains an impressive array of experiments which are based on sufficient treated animals and controls. The results as a whole would appear to show that phenothiazine is the best available anthelmintic for such important parasites as the stomach-worms of sheep, the strongyles of horses, and nodular worms in sheep and pigs.

As an example may be cited the group of experiments sponsored by the Agricultural Research Council, the results of which were published in 1940 as a symposium edited by Taylor & Sanderson (12, 23, 27, 30, 36, 44). Various workers in different parts of this country used in the aggregate some 150 horses, 300 to 400 sheep, 70 to 80 goats, and a few cattle, pigs, and dogs. The drug was found to be remarkably efficient against horse strongyles and the stomach-worms of ruminants, and in varying degree less efficient against other helminths. A good case of complete efficacy can be found on page 640 of the symposium, where Taylor & Sanderson show that five untreated control lambs contained from 2,100 to 55,400 stomach-worms whilst six lambs given 15 gm. of Swales' phenothiazine mixture contained no stomach-worms at all.

Beyond those already mentioned, there are at least 18 more papers relating to sheep and published in 1940. These include one in which Gordon (9) claims good results against immature *Haemonchus*, and another where phenothiazine is favourably compared with tetrachlorethylene and with copper and nicotine sulphates by Gordon & Whitten (11). A further 18 papers in 1941, dealing with sheep, include Holman & Pattison (20) describing a pathologically unimportant anæmia in sheep given phenothiazine at 0.75 gm. per Kg., Rowlands & Harbour (37) using the drug in cases of " pining ", and three more papers comparing it favourably with copper and nicotine sulphates (Boddie *et al.*, 1, Shorb *et al.*, 40, Stewart & Crofton, 41).

*Numbers in parentheses refer to the bibliography on pp. 55-56.

Nearly all the papers so far mentioned are strongly favourable to phenothiazine. But in 1941 a few discordant notes are sounded. Toxic symptoms are reported in horses by Errington (4) and by Habermann *et al.* (14) in the U.S.A., and in cattle and pigs by Roberts (35) in Australia. Deaths in horses, ascribed to phenothiazine at least in part, are recorded by Hatcher (19), by Schmidt *et al.* (38), and by Wolfe & Dennis (45), all in the U.S.A. Apart from toxicity, failure as an anthelmintic is claimed by Shaw (39) who used the drug in 72 Oregon sheep. The same is true of the papers emanating from the Institute of Agricultural Parasitology, St. Albans (25, 26, 31, 32). The workers here were concerned mainly with the more general problem of how an anthelmintic experiment can best be designed, executed, and assessed, but, using phenothiazine in their tests, they reported failures with it at doses of about 5 gm., 10 gm. and 30 gm. per lamb, and success at 40 gm. At the first two dosage rates the drug was given as a drench, with and without added wetting agents, and at 30 gm. the tablets used were unsatisfactory, probably through age. They considered the nature of the wetting agent to be crucial and suggested that only freshly made tablets are serviceable.

Of 11 papers relating to sheep and published in 1942, two refer to the incorporation of phenothiazine in licks (Habermann & Shorb, 15) or in concentrates (McCulloch, 27). Swales *et al.* (43), summarizing their experience in Canada, claim that *Oesophagostomum columbianum* has been practically eradicated as a source of disease by doses of 40 gm. in adults and 30 gm. in yearlings. Mönnig (29), in the official journal of the South African Dept. of Agriculture and Forestry, states that " Tests . . . undertaken at Onderstepoort have given results which were not so striking [as those reported elsewhere] and they did not outweigh the disadvantages of the remedy, except in the case of certain worms of horses ".

From the first, methods of administering the large quantities of the drug required have varied greatly. Thus, Gordon (6), gave it as a drench, and also (7, 10) in gelatine capsules. Roberts (34), used a suspension in 4 per cent. copper sulphate solution, or in paraffin. Swales (42) devised his explosive tablet. Harwood *et al.* (16) mixed the dry powder with grain and molasses, but found that the mixture was not always readily eaten.

The dosage has also varied greatly. In the first anthelmintic paper Harwood *et al.* (18) used as much as 1 gm. per Kg. of live weight. In subsequent work, both low and high doses have been tried; Gordon (7) used 0.2 gm./Kg.; Peters *et al.* (32), comparing 0.15 with 0.3 gm./Kg., found the former as effective as the latter in reducing egg counts (neither dose reduced worm counts). At the other end of the scale Swales (42), using 0.66 to 1.1 gm./Kg., has declared himself opposed to inadequate doses. Where the dose is given on a per capita basis, the general range is from 4 gm. (Roberts, 34) or 5 gm. (Lapage, 24) to 40 gm. (Peters *et al.*, 31) or, as a test of tolerance, in two 86-89 lb. lambs, 400 gm. (Taylor & Sanderson, 44).

Various criteria of efficacy have been adopted. Broadly, the Australian and American workers appear to favour Hall's " critical test " (e.g., Harwood *et al.*, 16) combined or not with egg counts before and after treatment. In the " critical test " one counts worms passed in the fæces after dosing and compares the numbers with those found *in situ* at post-mortem. It has the disadvantage that small worms may be digested before being voided and, in the absence of controls, disregards the normal voiding of large numbers of worms at certain seasons. More usual in this country is the controlled test, comparing post-mortem worm counts from treated and untreated animals. Counts of worm eggs, usually by a " dilution " (as opposed to a " concentration ") technique, have the advantage that they can be made both before and after treatment, but they are subject to very considerable variation. The more practical criterion is the differential growth of treated and untreated animals, as measured by weighing before and after treatment. J. Leiper & Peters (25) have claimed that phenothiazine has no effect on

the weights of worm-free sheep and goats, so that weights can properly be regarded as a criterion of anthelmintic action.

All these methods yield data which should enable efficacy to be assessed with some precision. That this is seldom done in actual fact is probably due to the essentially statistical problem of variation, which leads most workers to present their data and to rely on common sense in making a rough evaluation of them. The St. Albans workers (25, 26, 31, 32) have been largely concerned with measuring and making allowance for the observed variations.

## 2.—The Present Series of Trials

### (a) *General Plan*

In the preliminary survey of the problem, in both its clinical and its practical aspects, it was felt that the discrepancies in the findings of different workers using phenothiazine were due, at least in part, to their having had different aims, and having tested the efficacy of the drug by different criteria of success. It will appear from the results consistently found at the four centres carrying out the present series of experiments that this circumstance, in conjunction with frequent differences in the level of dosage tested, is capable of explaining very wide discrepancies. There are, in addition, the differences ordinarily existing between different batches of experimental material, such as, in the present case, differences in age of lambs tested, or differences in breed and in previous treatment.

There are three observable characteristics of treated or untreated sheep by which the efficacy of treatment is commonly assessed from different standpoints, as follows:—

(i) *Counts of Eggs in Fæcal Matter.*—The technique for the evaluation of these is fully set out in Section 2j (p. 11). This observation has the great advantage that it can be carried out on living animals, and therefore is of value not only after a period of treatment as a means of gauging its efficacy, but also as a preliminary observation before treatment has been commenced as a means of measuring the degree of infestation with which each lamb enters the experiment. It is also capable of relatively great quantitative precision. The diverse species of parasitic worms are not ordinarily differentiated in the egg counts. The counts taken at the four co-operating centres distinguished the eggs of *Strongyloides* and *Nematodirus,* and a third group comprising the total of the eggs counted from other species, excluding *Trichuris* and *Moniezia.*

(ii) *Worm Counts.*—After slaughter direct counts, or sampling estimates, were made of the numbers of adult worms, distinguished at least as far as genera, found in the fourth stomach or in the small or large intestine. The worm-counting technique is described in Section 2k (p. 12). Both qualitatively and quantitativly such counts afford a better assessment of the intensity of infestation than can be afforded by egg counts. They can, however, be taken only on slaughtered lambs and cannot, for this reason, be used to assess the initial degree of infestation. Moreover, counts of individual species vary enormously from lamb to lamb.

(iii) *Live weights.*—From the point of view of practical application, in animal husbandry, the progress of the animal in growth, as measured by live weight, is of an importance superior to that obtainable by any purely clinical observation, for it affords a measure of the net effect of treatment on the commercial value of the animal. Since, apart from the effect of drugs upon parasitic species, the drugs may be presumed to have (and, in fact, show evidence of having) direct though perhaps temporary effects upon the growth of the lambs, it is essential that the net consequences of treatment through all chains of causation, explored or unexplored, should be assessed in determining its practical value.

Broadly speaking, in the present trials information was required on two different topics: (i) the effects of the drug on particular species of worms, and (ii) the resulting net effect upon the lambs themselves. Light on the former and more academic topic would be shed by counts of eggs and worms; the problem of re-infestation, inevitable in a flock at pasture, would be solved by keeping the lambs inside, and the experiment need last only a few weeks. On the other hand, the second and more practical topic would best be served by the periodical weighing of lambs over a period of many weeks, the lambs being kept at pasture under natural conditions.

(b) *The Drug*

Having in mind the discrepant results obtained in certain of the earlier trials, and the variations in colour and texture which are met with in different commercial samples of phenothiazine, it was regarded as important to work with a sample of the drug, the chemical and physical properties of which were clearly established.

An amount sufficient for the whole series of trials was kindly supplied by Imperial Chemical Industries Ltd., to whom we should wish to express our thanks. Its composition and characters were as follows:—

| | |
|---|---|
| Phenothiazine ... ... ... ... ... | 98 per cent. |
| Moisture ... ... ... ... ... ... | 2 per cent. |
| Crystallizing Point ... ... ... ... | 182° C. |
| Arsenic ... ... ... ... ... ... | Less than 1 p.p.m. |
| Lead ... ... ... ... ... ... | Not exceeding 3 p.p.m. |
| Copper ... ... ... ... ... ... | Not exceeding 2 p.p.m. |
| Colour of 2 per cent. solution in acetone ... | Very pale olive green |

It thus fulfilled the specification suggested by Imperial Chemical Industries as desirable for phenothiazine for veterinary use:—

> A pale olive green to grey-green powder of soft texture, giving in 2 per cent. solution in acetone a clear and bright solution not darker than pale olive green, and containing not more than a trace of insoluble matter, the powder melting at not less than 181°C.

As phenothiazine itself is not easily wettable with aqueous fluids, it was converted into a wettable preparation by mixing intimately with sodium cetyl sulphate, which is a very effective wetting agent. The preparation, as supplied to us, contained 96 per cent. of phenothiazine and 1.25 per cent. of sodium cetyl sulphate, the balance being moisture and traces of inorganic salts.

(c) *Method of Administration*

Phenothiazine may be administered as a drench, or in the form of tablets. Since both tablets and drenches are used for this purpose, in ordinary practice, it was decided, in the present series of trials, to compare the efficacy of the drug given in both forms.

In the case of the tablets, sodium cetyl sulphate was used in the same ratio as in the drench, together with a small amount of starch as binding agent. The tablets were in the form of cylinders with rounded ends, were $1\frac{3}{8}$ in. long by $\frac{1}{2}$ in. diameter, and weighed approximately 5.45 gm. Each tablet contained 5 gm. phenothiazine and 0.08 gm. sodium cetyl sulphate, the balance being starch. Owing to the presence of starch and wetting agent, the tablets rapidly disintegrated and dispersed when added to water.

(d) *Dosage*

Previous reports have indicated that the egg count may be decreased by quite small doses of phenathiazine, without any significant increase in the animal's weight, while other reports have suggested that growth rate is favourably affected by larger doses. It was, therefore, felt desirable to test a wide range of dosage.

There are certain advantages in adjusting dose to body weight; but tablets are not easily broken into fractions of any desired weight, and it was, therefore, decided to give each lamb in any one dose-group a fixed weight of phenothiazine, each dose being a multiple of 5 gm., the amount contained in a single tablet. The range of dosage selected was: 0 gm. (controls), 5 gm., 10 gm., 20 gm., 30 gm., 40 gm., and 50 gm., per lamb.

At each centre a " unit flock " contained 28 lambs. Two lambs received nothing, and two received the same amount of sodium cetyl sulphate, as a drench, as was present in the 50 gm. dose. These four lambs served as controls to all treated groups, taken as a whole. When lambs receiving tablets were considered separately, the two lambs receiving nothing were taken as controls, while the two lambs receiving the sodium cetyl sulphate were treated as controls for the lambs receiving phenothiazine as a drench. The remaining 24 lambs in each unit flock were divided into six groups of four lambs each. In each group two lambs received the appropriate dose as a drench, and two as tablets.

At St. Albans and at Edinburgh one unit flock was housed in suitable buildings for about six weeks, so that re-infestation was prevented, and a second unit flock was maintained on pasture for about 16 weeks. The flocks kept under these two sets of conditions are referred to as " inside " and " outside " flocks. At Cambridge, one inside flock was observed; at Weybridge one inside and four outside flocks. Thus, in all, this series of observations covered four inside flocks and six outside flocks, a total of 280 lambs.

(e) *Condition of Lambs*

In selecting the lambs for these trials, it was felt that animals showing obvious signs of acute parasitic gastritis might be unsatisfactory, since many of them might die in spite of treatment. On the other hand, too lightly infested lambs would not provide satisfactory material for assessing the fall in egg or worm count in relation to dose, nor would they, from the practical point of view, represent the type of infected animal most in need of treatment. Each centre, therefore, endeavoured to buy the necessary number of lambs with a level of infestation of about 1,000 eggs per gramme of fæces. The breeds used at each centre are specified below in Section 3.

In order to include at each centre a few spares, which could be used for replacement if need arose, 60 lambs were purchased to provide 56 animals required for two unit flocks.

(f) *Randomization*

For each centre a separate set of 60 random numbers was provided, so that any one lamb of the 60 was allotted to its flock (inside or out), to its dosage level, and to tablets or drench, purely by chance. Lambs were ear-tagged with the numbers 1 to 60, and it made no difference whether this tagging was strictly random or (say) in order of size; randomization was effectively secured by the set of numbers prescribed.

(g) *Management of Lambs*

To prevent re-infestation among the inside flocks, their pens were thoroughly cleaned out twice each week. The ration supplied was, per lamb per day: 3 lb. mixed hay and oat or wheat straw, $\frac{1}{2}$ lb. crushed oats, $\frac{1}{2}$ lb. bran. The same ration was given to the outside flocks later in the year if and when the pasturage failed. Since the artificiality of inside conditions might lead to a temporary setback, these lambs were kept inside for at least a week before the experiment started so that they might become acclimatized.

Centralized analysis of all the data necessitated a uniform system of recording weights, egg, and worm counts; suitable record forms were therefore circulated to all centres.

(h) *Time-table of Experiments*

In both outside and inside flocks the lambs were weighed immediately before dosing and again at the end of the experiment. In addition, the outside lambs were weighed four times during its course, the interval between any two successive weighings being three weeks. Similarly, in both flocks, eggs were counted on three separate days before dosing and on six further days at the end of the experiment. The inside lambs were killed as soon as convenient during the week following the last egg count, the contents of abomasum and small and large intestine being separately preserved for subsequent worm counts.

The time-table reproduced at Appendix I is self-explanatory; it was designed to avoid handling both flocks on any one day. Each centre proceeded independently in securing the requisite lambs and in the absolute dates of starting the experiments, but all conformed as to relative time-table and technical details.

(i) *Weighing Lambs*

Most weighbridges made to take sheep are capable of weighing to the nearest $\frac{1}{4}$ lb. In such cases weights were recorded in $\frac{1}{4}$ lb. units, and at any one centre lambs were weighed at the same time of day on each occasion.

(j) *Counting Eggs*

The following simplification of the McMaster egg counting technique, known to produce counts in close agreement with the theoretical Poisson distribution, was adopted for use in these trials.

A sample of fæces was weighed on a paper disc and transferred to a 100 ml. cylinder containing 30 ml. of water. The fæces were broken up (after soaking 5 minutes if hard) by a plunger consisting of a disc of perforated zinc slightly smaller than the diameter of the cylinder and secured to a stout wire handle. The mixture was made up to 60 ml. with saturated salt solution, the plunger was moved rapidly up and down keeping the disc below the surface of the suspension, and after 40 strokes a pipette-full of suspension was withdrawn and one cell of a McMaster slide filled. Plunging continuing, the two other cells were similarly and separately filled. The slide was left two minutes for eggs to float. If 2 gm. of fæces are weighed, the total count from three cells multiplied by 66.6 will give eggs per gramme of fæces; however, the actual counts per cell, and not eggs per gramme, were recorded.

The essence of the McMaster (Gordon & Whitlock) technique is that one counts the eggs lying under a centimetre square engraved on the underside of a coverglass which is supported 0.15 cm. above a slide by means of cemented glass slips. Thus, the volume of fæcal emulsion examined in each cell of the slide is 0.15 cc. Since the suspending liquid is a half-saturated solution of salt, the eggs float up into one optical plane immediately below the coverglass and can readily be counted. The original McMaster slide had three such cells in line, the three squares being engraved on a single coverglass 3 in. long. Accurate counting necessitates the use of a mechanical stage, and many of these have not sufficient lateral travel to cover the three cells. Two- and three-cell slides were used in the present experiments. The mechanical stage scans the centimetre square in a series of about six parallel strips (depending on the optical equipment used); hence there is some danger of counting a single egg twice if it lies near the edge of a strip. This has been obviated by having additional strip lines engraved within each square, the entire width of a strip falling within the field of vision. The lines of the square can be engraved mechanically with high precision, and the larger source of error in McMaster slides lies in the depth, where flatness of slide and coverglass, and thickness of supports and of cement, all contribute to variation. For the slides used in the present experiments a tolerance of 1 per cent. in the volume was aimed at.

| *Condition of Fæces* | *Dry Matter %* | *Weight of Fæces* (for the McMaster technique) |
|---|---|---|
| Formed pellets ... | 30 (up to 40) ... | 2 gm. |
| Soft pellets ... ... | 24 ... | 2.5 gm. |
| Soft : no pellets ... | 20 ... | 3 gm. |
| Mushy ... ... | 12 ... | 5 gm. |
| Liquid ... ... | 8 ... | 7.5 gm. |

This worked fairly well in practice, apart from occasional unexplained anomalies.

The inside flock made reasonable progress; there were no casualties and the flock gained in weight an average of about 1 lb. per head per week during the six weeks of the experiment. Half the lambs, one from each pair of replicates, were slaughtered and the carcases dressed at Winches Farm on the 10th November, and the remainder on the 13th. As soon as the butcher slit open the abdomen, ligatures were tied above and below the abomasum and at the ileo-cæcal valve. No livers showed gross signs of fluke (no eggs had been found) but a moderate lung-worm infestation was present in all 28 lambs.

The outside flock also made fair progress at first. During the nine weeks separating the first and fourth weighings they also gained about 1 lb. per head per week. But, on the 29th October, shortly after the last signs of fly had disappeared, foot-rot was detected. In spite of frequent applications of copper sulphate solution and the paring of hooves, the infection rapidly spread through the whole flock. At its height, a few lambs were unable to stand. However, constant treatment with change of pasture saved the situation and no lambs were lost; by the 5th December the last traces had gone. But the lambs had received a check, and over the 15 weeks from first to last weighings the net average gain was under 6½ lb. per head. Throughout the experiment this flock was on permanent grass, their movements from field to field being as follows:—

| *Field* | *Date admitted* | *Acreage* |
|---|---|---|
| D3 and 4 | 16.9.41 | 0.99 |
| A3, 4, 5 | 3.10.41 | 1.24 |
| A2 | 24.10.41 | 1.72 |
| F5 | 14.11.41 | 1.33 |

Early in December the grass began to fail and from the 9th December until the experiment finished on the 16th January, 1942, the 28 lambs were given a daily supplement of 10 lb. crushed oats and 10 lb. bran. They were sold alive at the local market, fetching a poor price.

(b) *University of Edinburgh, and Animal Diseases Research Association, Moredun*

The flock used at Edinburgh consisted of 70 Cheviot hill lambs about six months old, obtained from Eskdale, Dumfriesshire. The animals were brought to the Moredun Institute on August 22nd and were ear-tagged the following day. Preliminary egg-counts gave a range of variation of from 100 to 4,000 eggs per gramme, with an average of about 1,000 e.p.g.; but, as an appreciable number of lambs had counts below the desired minimum of 1,000 e.p.g., it was decided to place the lambs on a selected heavily contaminated pasture at the Institute in order to increase the infestation. The animals were left on this pasture for four weeks and were then brought in and randomized, the "inside" group being placed in seven separate loose-boxes, each box containing the four lambs of the same dosage group. The "outside" group was placed in a five-acre paddock of old pasture which had been grazed continuously with horses since 1934, except for four months from December, 1939, when some sheep were on the ground. Egg counts taken on 29th September, when the experiment started, showed that the average level had increased to about 2,500 e.p.g.

As 56 lambs were required for the experiment, this left a surplus from the 70 originally bought. Part of this was used by H. H. Holman & I. H. Pattison for observations on the blood-picture of sheep treated with phenothiazine (1942, *Vet. Rec.*, **54**, 215-216). There remained four, which were used as additional controls to the " outside " flock and which are recorded with the controls in Appendix IV of this Report.

Tablets were administered by means of a gag and large forceps, and considerable difficulty was encountered, particularly at the higher dosage rates, owing to the small size of the lambs. After the fatal asphyxiation of one lamb, it was decided to rest each animal when three or four tablets had been given, and to complete the dose after an interval. The drench was administered without difficulty from a long-necked bottle.

With regard to egg counting technique, no attempt was made to estimate dry-matter or to grade the fæces by appearance. Very few liquid or mushy samples were encountered and even when they did occur, the storage of the fæces in newspaper bags in a refrigerator over-night resulted in the consolidation of the samples by the absorption of excess moisture.

Despite the " acclimatization " of the " inside " group for a week before the experiment began, the animals did not take readily to inside feeding and it was not until the third week of the test that the lambs were feeding normally. By the end of the experiment, however, with two exceptions, all had increased in weight. In the " inside " flock, one lamb died, showing on post-mortem examination advanced necrosis of the liver, probably caused initially by fluke infestation.

The feeding of the " outside " flock presented no difficulty. The lambs, however, refused to eat hay, probably on account of the fact that a fair bite of grass was available on the paddock throughout the period of the experiment. The prescribed ration of bran and oats was, however, readily taken during the latter part of the experiment. One lamb from this group died, but no obvious cause of death could be ascertained.

The " outside " animals were slaughtered at the end of 16 weeks and, like the lambs of the " inside " group, were sold in the dead meat market. Even the best of them could hardly be considered to be above store condition and this would account for the low grade in which they were placed. The slaughtering of the " outside " group afforded the opportunity of collecting the intestinal contents and making worm counts, as was done with the " inside " group. The worm counts have been submitted for statistical analysis, but the results are not included in this Report, as the work was outside the prescribed scheme.

(c) *Institute of Animal Pathology, Cambridge*

After considerable difficulty two lots of lambs were found in Norfolk which had the average egg count of 1,000 e.p.g. or over required by the plan of the experiment. The preliminary single egg counts suggested that *Nematodirus* and *Strongyloides*, both of which would be affected little or not at all by phenothiazine, were probably numerous. The eggs of *Strongyloides* comprised about half the total egg count of some of them. After further egg counts an experimental flock was made up of lambs which all showed, before the experiment began on November 24th, 1941, egg counts of 1,000 e.p.g. or over on two out of three successive days. The lambs were Suffolks and Suffolk-Hampshire crosses aged 6-9 months. This flock was randomized and put in an enclosed compound, which had a concrete floor and sheds on two of its sides for shelter and fæcal sampling.

The prescribed time-table and technique were followed throughout. Usually all the figures and calculations were checked by at least two workers. By agreement dry-weight estimations were done on dosage groups 5 g. and 30 g. Foot-rot developed in some of the lambs, but this was never serious.

Two lambs died during the experiment. One of these, No. 56, resisted the administration of its dose of 10 tablets. It died on 4th December, 1941, six days after the dose. Autopsy revealed a gangrenous ulcer of the soft palate, due to trauma by the forceps used for giving the tablets. There was a massive inhalation pneumonia. Lungworms (*Dictyocaulus filaria*) were present, but were not causing acute inflammation. The pleuræ were acutely inflamed and there were about 300 ml. of fluid in each pleural cavity. The small intestine contained about 1,600 *Nematodirus* and the large intestine 12 *Trichuris*. No other nematodes were found. The other lamb, No. 47, began to scour soon after the beginning of the experiment; it died on 26th December, 1941, the day of the last egg count. At autopsy a confluent broncho-pneumonia was found, which was almost certainly due to the inhalation of foreign substances, although phenothiazine could not be identified in the lungs. All other organs were normal and very few nematodes were present. It is possible that the scouring of this lamb was due to some non-helminthological condition, for the detection of which there were no provisions in the plan of the experiment.

The lambs were slaughtered at a Cambridge abattoir, where, immediately after the abdomen had been opened and before the animals were hung up, ligatures were applied between the omasum and the abomasum, at the pylorus, at the ileo-cæcal junction and as near the anus as possible. The digestive tracts were then brought to the laboratory and the worms in them were counted by the prescribed technique.

(d) *Ministry of Agriculture and Fisheries Veterinary Laboratory, Weybridge*

*Inside Flock.*—Only two of these 28 sheep were showing a high egg count at the commencement of the experiment. At randomization one of them fell into the control group and the other was given a 20-gm. dose in tablet form. The control animal succumbed during the third week. The dosed animal, although remaining in very poor condition, remained alive throughout the trial.

*Outside Flocks.*—Four groups of sheep were treated experimentally by this centre, all of them remaining on the farms of their owners. Flocks were selected which although carrying a fair worm burden, as shown by egg counts, did not show clinical symptoms of parasitic gastritis apart from general debility. It was hoped to test the value of phenothiazine as a preventive measure, in these flocks, before the onset of severe symptoms of parasitic gastritis and to ascertain whether the poor thriving often seen in autumn lambs would respond to treatment with this drug. By chance groups B and D were on farms where the general standard of management was good and store animals received a certain amount of supplementary food, whereas groups A and C were maintained on a lower plane of nutrition.

In Group D the experimental details were carried out exactly as for the outside flocks at the other centres. In the three groups A, B and C, however, only three egg counts instead of six were made at the end of the experiment.

*Group A.*—These lambs were in poor condition at the beginning of the trial, several of them coughing from lung-worm infestation, but three weeks after dosing the shepherd remarked on the noticeable improvement in the general appearance of the flock. At this time the pasture, although of poor quality, was plentiful; in eleven weeks' time, however, it was becoming scarce, and by the thirteenth week three deaths had occurred for which the shepherd blamed lack of food. No extra food was allowed by the farmer and the sheep were kept on the same eleven-acre pasture the entire time. Coughing had ceased by the end of the experiment. These were South Down × Kent lambs of about six months old.

*Group B.*—At the beginning of the experiment these lambs were in good condition. They were kept on the same 15-acre field, running with a few cattle until the thirteenth week, when they were brought nearer to the farm and given supplementary food, because

of snowy conditions. One sheep was lost during this period. The farmer commented on the improvement in the flock during the experimental period. These were Kent lambs born in March and April.

*Group C* had formed part of a flock of late lambs, the majority of which were in very poor condition. A few were coughing from lung-worm infestation. This group was moved constantly during the observational period to supplement the poor pasture by giving them access to cabbages, and later to swedes. They also received cut mangolds during a period of heavy frost and snow from the eighth to the twelfth weeks of the experiments, but frequently these were not eaten on account of being frozen. The condition of the groups was still very poor at the end in spite of the improvement actually shown in the analysis of the results. A post-mortem examination was made on one of the sheep receiving a 40-gm. dose (tablets) at the end of the trial, as this animal appeared in particularly bad condition. It was found to be suffering from a very heavy lung-worm infestation. This examination was useful, as it indicated that the flock was carrying a multiple infestation of unusual variety: *Trichostrongylus axei, T. vitrinus, Ostertagia, Haemonchus, Cooperia, Nematodirus, Bunostomum, Strongyloides, Moniezia, Oesophagostomum* and *Trichuris*. These lambs were a mixed group of 15 Devon Closewools, 4 Exmoor Horns, and 9 Blackfaces, all spring born.

*Group D.*—These lambs were in good condition apart from several which had been fly struck. This cleared up before the second weighing with the exception of one animal in the control group, which had to be killed by the shepherd during the sixth week. The farmer expressed satisfaction with the progress of the flock. They were kept on their original pasture until the thirteenth week, when they were moved to a fresh clover ley. They were South Down × Kent lambs born in March and April.

*Method of Dosing*

Dosing was carried out under normal farm conditions, i.e. in badly lighted sheds with only buckets of water provided. A small proprietary balling gun was used, the tablet being held by a piece of thick rubber tubing, of diameter sufficiently small to hold the tablet, but not too small to prevent its easy release. In the five groups of animals treated only one individual showed any difficulty in swallowing the large numbers of tablets necessary. The phenothiazine liquid was given by the usual dosing syringe, but it was found that the density of the liquid obscured the graduation of the measuring utensils, which necessitated constant rinsing in fresh water.

## 4.—Statistical Analysis of Results

The actual live-weights and egg and worm counts are set out in the Appendices II, III and IV. Succeeding appendices, for which R. A. Fisher and B. G. Peters are responsible, give the technical details of the various methods of statistical analysis used. The general outcome of the trials, as assessed from these analyses, is discussed in the following pages, in the order: egg counts, worm counts, live weights, with a final comprehensive section.

### (a) *The Effect of Phenothiazine in Reducing Total Egg Density*

The initial and final egg counts on the four inside flocks which were maintained for six weeks under conditions which prevented re-infestation, provide the best data for assessing the effect of varying the dose of phenothiazine on the change in the fæcal egg count which results from treatment. Some previous workers have relied on a simple comparison between the final egg counts in treated and untreated groups, but this takes no account of the initial level of infestation which may vary widely.

Appendix II shows the total number of eggs counted from each lamb of the inside flocks. The values under the main heading " Initial Egg Counts " are based on the sum of three counts made on three different days during the week before treatment.

Each value under " Final Egg Counts " is the sum of six counts made during the third and fourth weeks after treatment. In order that the counts should be comparable, the sum of the three initial egg counts has in each case been multiplied by two. This summation of respectively three and six counts tends to smooth out the day-to-day variation, which is considerable.

A comparison of the initial and final total egg counts for each dosage group of four lambs shows that treatment has had considerable effect, but two circumstances rob these totals of precision. First, since the final counts from the four lambs at any one dosage show marked random variation among themselves, it follows that the differences between the totals will be partly due to this random variation. Secondly, the lambs did not enter the experiment all showing the same egg density, so that the initial egg counts must be allowed to influence any assessment of treatment differences in the final totals.

As the lambs were assigned to treatments by a strictly random process, it will be permissible to apply to these data standard statistical techniques, which themselves presuppose such a random assignment. In particular, the two sources of confusion just mentioned can be dealt with by two statistical procedures (in actual computation the two are combined): (i) The procedure known as " Analysis of Variance " enables the total variation to be split into a part due to treatment and a part due to mere random variation; if the first part is sufficiently greater than the second, the effect of treatment can be judged to be real by a test of significance of any chosen degree of severity. (ii) The calculation of " Regression " measures the average dependence of final on initial egg counts, so that, with a given initial count, it is possible to predict what the final count should be, within limits of random variation, and supposing that treatment has had no effect. If, in any treatment group, the final count differs sufficiently from its predicted value, the effect of treatment will be judged to be real.

The results of applying this method to the data contained in Appendix II are set out in Table 1.* In this comparison both the lambs treated by drench and those treated by tablets are included.

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* If details of the appropriate statistical computation are required, they will be found in Fisher's " Statistical Methods " (5) Section 49.1 (" The Analysis of Covariance "). There is one complicating feature of the present analysis, however, which can best be explained by contrasting with it the analysis appropriate to (say) weights of lambs. If any two lambs under the same treatment entered the experiment with a difference in weight of (say) 5 lb., it is reasonable to expect that roughly the same difference would be found at the end of the experiment. To such a linear relationship the simple form of the regression equation is applicable :

$$y = c + bx$$

where $y$ is final weight, $x$ initial weight, $b$ the regression coefficient, and $c$ a small constant. Also, the " adjusted " final weight (adjusted, that is to say, so as to take proper account of initial weight) is given by the difference :

$$(y - bx)$$

On the other hand, in the case of egg counts where there are very large reductions from initial to final counts, it is inherently unreasonable to expect such a simple linear relationship. If any two lambs under the same treatment entered the experiment with egg counts of $x_1$ and $x_2$, it is more reasonable to suppose that the ratio $x_1/x_2$ would remain roughly the same at the end, than that the difference $(x_1 - x_2)$ would do so. In such a case the relationship is logarithmic ; the equation becomes :

$$\log y = c + b \log x$$

and the adjusted final counts are given by the ratio : $y/x^b$

So that, with any two lambs treated alike, one would expect the ratio of their final counts to be roughly the same as that of their initial counts. Actually, the initial counts are liable to error and the counts will naturally vary in the course of time from various causes, so that the ratio will probably change. In the event, comparisons made within the same inside flock showed generally that the ratio appeared to diminish. Each flock was used to ascertain how much lower in fact the final ratios were compared with the initial ratios. Thus, at Edinburgh, the final ratios were less in the ratio 0.64. Once this had been determined, proportional expected final counts were ascertainable for each lamb in the flock. These were compared group by group with the final counts observed (using for comparison the ratio of observed to expected final counts), taking the control or untreated group as 100.

TABLE 1

RATIO OF OBSERVED TO EXPECTED FINAL EGG COUNTS : INSIDE FLOCKS

| *Dosage gm. per lamb* | *Edinburgh* | *St. Albans* | *Weybridge* | *Cambridge* | *Average* |
|---|---|---|---|---|---|
| 0 | 100 | 100 | 100 | 100 | 100 |
| 5 | 35 | 42 | 42 | 52 | 43 |
| 10 | 17 | 59 | 34 | 15 | 31 |
| 20 | 17 | 9 | 48 | 49 | 31 |
| 30 | 17 | 33 | 24 | 33 | 27 |
| 40 | 9 | 13 | 15 | 82 | 30 |
| 50 | 9 | 18 | 15 | 51 | 23 |

It will be seen that, at all centres and at all doses (twenty-four comparisons in all) the final egg count is diminished in comparison with the untreated lambs. Secondly, the greater part of the diminution has been brought about by the first 5 gm. of the dose. Evidently extremely small doses are effective in diminishing the total egg count. Beyond this level the diminution is comparatively small, and in most flocks too irregular to be detected on the values for only four lambs. On the average of four flocks, however, the 50 gm. counts give the lowest figure, though beyond 10 gm. the changes are slightly irregular. The Standard Error for the average of four flocks at any given dosage is about ± 19 per cent. of each value shown.

It will appear from the worm counts (Appendix III) that different species differ greatly in susceptibility to the drug. The most susceptible species, *Haemonchus contortus*, is believed to contribute largely to the egg count, while some of the less susceptible may contribute relatively few eggs. This consideration may be thought sufficient to explain the high apparent efficacy of small doses, as also the fact that the response as measured by egg count is apparently greater in some flocks than in others, at Edinburgh, for example, than at Cambridge. The response at Edinburgh was so regular that an attempt was made to compare the actual counts with a series of expectations based on formulæ commonly used in toxicology to compare the quantitative effects of various dosages. This comparison is set out in Appendix VI.

Turning to the outside flocks, and comparing the initial and final egg counts by the same method, we obtain the figures set out in Table 2.

TABLE 2

RATIO OF OBSERVED TO EXPECTED FINAL EGG COUNTS : OUTSIDE FLOCKS

| *Dosage gm. per lamb* | *Edinburgh** | *St. Albans** | *Weybridge* | | | |
|---|---|---|---|---|---|---|
| | | | *A*† | *B*† | *C*† | *D** |
| 0 | 100 | 100 | 100 | 100 | 100 | 100 |
| 5 | 43 | 72 | 211 | 102 | 28 | 785 |
| 10 | 51 | 110 | 102 | 91 | 19 | 213 |
| 20 | 36 | 97 | 108 | 98 | 15 | 213 |
| 30 | 42 | 113 | 77 | 254 | 4 | 200 |
| 40 | 42 | 104 | 67 | 94 | 31 | 181 |
| 50 | 25 | 81 | 242 | 69 | 6 | 337 |

*Based on six samples. †Based on three samples.

Since these flocks were maintained for about sixteen weeks under conditions that permitted re-infestation, we should not expect the diminution in fæcal egg count to be so large as in the inside flocks, in which re-infestation was prevented. Moreover, since the risk of re-infestation probably varied from one centre to another, we should expect the figures from the different centres to be less uniform. Both these expectations are realized.

It will be seen that in the case of St. Albans and Weybridge A, B, and D, any reduction in egg count due to phenothiazine has disappeared, owing to re-infestation during the 15 weeks after dosing. The extraordinarily high values at Weybridge D are due to the circumstance that the group at zero dosage (which controls the values of all other groups) was by chance the least infested at the end of the experiment. At Edinburgh and Weybridge C, however, the egg counts continue to show marked effects of dosage. The outside flock " Weybridge C " is not strictly comparable with the Weybridge inside flock, but the two Edinburgh flocks were random selections from a single group of lambs, and the Edinburgh data from Tables 1 and 2 show that the reduction after 15 weeks, while far less than after five weeks, is nevertheless marked. This suggests that these two flocks were kept on pastures relatively free from infective larvæ. The evidence in respect of Weybridge C is not conclusive; the lambs were maintained after dosing on a semi-arable system but with some access to permanent pasture. At Edinburgh the case is clear. The experimental lambs were grazed on a pasture which had carried sheep for four months from December, 1939, but apart from this had been grazed exclusively by horses from 1934 until the date of the experiment; this involves not merely the negative factor that sheep helminths were largely absent but also the positive cleaning effect of horses grazing for over a year after the prior contamination with sheep helminths.

With such wide discrepancies between the different outside flocks, no useful purpose would be served by calculating average figures.

(b) *Worm Counts*

The actual worm counts from individual lambs of the inside flocks are set out in Appendix III. No worm counts were made on the outside flocks. It is difficult to achieve any satisfactory statistical summary of such material. The frequencies for any one species of worm differ so greatly from lamb to lamb that no uniformity is to be expected from totals or averages. Moreover, the measure of initial infestation supplied by the egg counts refers to a mixture of many species, and the attempt made with respect to *Nematodirus* to improve the comparison by this means failed through the relatively small numbers of eggs counted from this species.

The most that can be done is to pool the data from the four centres and present them in graphical form. While a logarithmic scale would be appropriate to these counts the difficulty arises that a zero count would be represented logarithmically by minus infinity. This can be circumvented by adding one unit to each actual count, plotting graphically log $(1+x)$; the logarithm of a zero count will then be zero.*

Figures 1-12 show graphs of log $a(1+x)$ plotted against dosage, using pooled data but drawing separate curves for the " Tablets " and " Drench " groups.

Surveying the data with respect to both frequency and degree of infestation, it is possible to classify the worms in order of decreasing susceptibility, as in Table 3. The

* A further difficulty is presented by the " small " worms which were estimated by sampling 10, 20, 30, or 40 ml. of intestinal contents from a total of 1,000 ml., thus giving dilution factors of 100, 50, 33.3, or 25 respectively. The counts of small worms included in Appendix III represent sample counts, $x$ multiplied by the appropriate dilution factor $a$. Clearly the unit should be added to the sample count, giving a logarithmic total count of log $[a(1+x)]$, rather than to the estimated total which would give a logarithmic total count of log $(1+ax)$. Thus, for example, while a zero count for *Haemonchus* means what it appears to mean, an *Ostertagia* count of zero per 40 ml. has a less precise meaning. It can be shown theoretically that, with an actual concentration of 75 worms per litre, 5 per cent. of all 40 ml. samples will contain no worms ; so that a count of zero per 40 ml. might well be consistent with a total lying between 0 and 75. With a count of zero per 40 ml. the expression $a(1+x)$ comes to 25 ; this is equivalent to a mean of one worm per 40 ml., at which concentration 37 per cent. of such counts will be zero. In such a case, $a(1+x) = 25$ is obviously a more reasonable estimate of the probable total than $1 + ax = 1$, or $ax = 0$. Similarly, with any low sample count greater than zero, the true value of the mean (of the population from which the sample is drawn) is more likely to lie above than below the value of the count, the disparity decreasing as the count increases.

data show that in all flocks *Haemonchus* is more completely eliminated by light doses than any other species. The two other parasites of the abomasum, *Ostertagia* and *Trichostrongylus axei*, are also susceptible to doses up to 20 gm., at which point half or more of the sheep are free from infestation.

TABLE 3

WORMS CLASSIFIED IN ORDER OF DECREASING SUSCEPTIBILITY TO PHENOTHIAZINE

| *Class* | *Site* | *Species* |
|---|---|---|
| A | Abomasum | *Haemonchus* |
| B | Abomasum | *Ostertagia, Trichostrongylus axei* |
| C | S. intest. | *Cooperia, Trichostrongylus* spp., *Bunostomum* |
| D | L. intest. | *Chabertia, Oesophagostomum venulosum* |
| E | S. intest. | *Strongyloides* |
| | L. intest. | *Trichuris* |
| F | S. intest. | *Nematodirus, Moniezia* |

Classes C and D resemble each other in that there appears to be elimination of worms progressively up to the highest dose, but in Class D the response is considerably less marked than in Class C; this may be associated with the fact that the members of Class D, *Chabertia* and *Oesophagostomum,* occur in the large intestine.

In Class F the tapeworm *Moniezia* appears to be distinctly more abundant among lambs heavily dosed than among controls or lambs lightly dosed. The same may be true of *Nematodirus,* the frequencies recorded for which are, however, exceptionally irregular—possibly owing to the fact that these worms tend to form dense clusters in the intestinal contents.

In Class E there is some apparent increase from 0 to 20 or 30 gm., with a decrease for higher doses. It appears possible that, owing to the removal of competing species, the parasites of Class F are able to maintain their infestation of lambs from which, without treatment, they would have been partly eliminated, and that doses of 20 or 30 gm. confer a similar benefit on the members of Class E. Of the 12 kinds mentioned, there can be little doubt that surviving adult parasites of the first eight are appreciably diminished, and that at the higher doses some progress has been made towards disinfestation from the two parasites of Class E also.

Attention is drawn to the fact that the vertical scale used on the graphs for " Small " worms is one half the scale used for " Large " (see Section 2k, p. 12).

The differences between the curves for tablets and drenches appear to be merely random, except in the case of the abomasal worms of Classes A and B, where there is a consisten suggestion that drenches are more effective than tablets; this point will be taken up again later.

(c) *Effect on Live Weight*

For a decision as to the value in farming practice of treatment with phenothiazine, reliance was placed on the weight increases of the outside flocks over a period of 15 weeks. There were six of these flocks, one each at Edinburgh and St. Albans and four more at farms near Weybridge, designated as Weybridge A, B, C, and D (see Appendix IV).

The final weight recorded for each lamb is, as might be expected, strongly associated with the weight at which it enters the experiment. In other material tested for shorter periods it has also been found to be sufficiently closely associated with the initial egg count to make it desirable to apply a correction for this factor also in calculating expected final weights to give added precision to the comparison. In the comparisons reported

here, therefore, account has been taken of both factors by the methods of multiple regression. The more technical data are in Appendix VII, and it will be sufficient here to point out one respect in which these comparisons differ from the egg count comparisons of Table 1.

In the case of egg counts, where the relationship was assumed to be logarithmic, the *ratios* of observed to expected final counts were expressed as *percentages* of the control ratios. In the present case, where the relationship is taken as being linear, the *differences* between observed and expected final weights are expressed as *differences* from the controls; i.e., the controls at each centre are made zero, and the resulting differences for the other six dosage groups are the values shown in Table 4, which includes data for the six flocks involved, and for the average of all, at each dosage level. The standard error for the average of six flocks at any one dosage level is just less than one pound live weight. From the nature of this table it follows that the actual values for each dosage level are relative to the control values; thus, the accident of abnormally low control values at Weybridge C has the effect of increasing all the other values at that centre.

While the individual figures based on only three or four sheep are somewhat irregular, at each centre the groups receiving 30 to 50 gm. all show an improvement compared with the controls. On the average of the six flocks the gains in live weight ascribable to treatment are seen to be large, and to increase rather regularly with dosage at least from 20 gm. onwards.

The comparisons available give a standard error of only $4\frac{1}{2}$ lb. for the individual lamb, which is a satisfactory level for practical experiments in animal husbandry.

TABLE 4

INCREASES IN LIVE WEIGHT COMPARED WITH UNTREATED LAMBS, IN LB. PER LAMB. OUTSIDE FLOCKS

| *Dosage in gm.* | *St. Albans* | *Edinburgh* | *Weybridge* | | | | *Average* |
|---|---|---|---|---|---|---|---|
| | | | *A* | *B* | *C* | *D* | |
| 5 | 2.51 | —1.07 | —3.99 | 1.03 | 8.07 | 1.04 | 1.26 |
| 10 | —1.70 | —4.68 | —3.63 | —0.29 | 10.74 | —1.11 | —0.11 |
| 20 | 3.60 | —0.28 | —0.54 | 5.89 | 8.69 | 4.43 | 3.63 |
| 30 | 1.35 | 4.65 | 4.31 | 2.13 | 9.78 | 1.28 | 3.92 |
| 40 | 7.78 | 1.25 | 1.65 | 5.35 | 7.92 | 1.54 | 4.25 |
| 50 | 6.08 | 2.90 | 1.57 | 5.49 | 12.34 | 4.68 | 5.51 |

In view of the possibility that these live weight data should favour particular dosage levels, each flock was tested for linearity of response in live weight to weight of drug administered; in no case were the departures from linearity statistically significant. The mean responses expressed in pounds live weight per 10 gm. unit of drug are indeed more concordant than could have been expected from the variations of individual sheep. These mean responses are as follows:—

| *St. Albans* | *Edinburgh* | *Weybridge* | | | |
|---|---|---|---|---|---|
| | | *A* | *B* | *C* | *D* |
| 1.38 | 0.88 | 1.04 | 1.15 | 1.19 | 0.73 |

The general average is 1.057 lb. live weight per 10 gm. phenothiazine, or approximately 48 times the weight of the drug, nearly independently of dosage within the range explored.

Since the different flocks differed undoubtedly both in degree and kind of infestation, the concordance of their average responses is an indication that similar responses could have been obtained in practical conditions from a very large number of commercial flocks.

The lambs in these flocks were selected as showing a level of infestation associated with about 1,000 eggs per gramme of fæces, and it might be expected that phenothiazine would produce greater gains in weight than in less heavily infested flocks, especially since it has been stated that it has no effect on the weights of worm-free sheep. Accordingly, it would be reasonable to suppose that the weight gains obtained in this experiment were rather better than those to be expected in the average flock. In order to test this point, it is necessary to investigate with the greatest possible precision what influence on final weight is exerted by initial egg count (as a measure of degree of infestation).

The above analysis took account of initial weight and initial egg count as independent variates, treating each flock separately, and the general effect was that the adjustments for initial egg count were small, making little difference to the final results. Much more elaborate calculations are required in order to do full justice to the possible advantage given by initial egg counts in improving the precision of weight comparisons. These calculations are detailed in Appendix VII, where four independent variates are analyzed, in respect of both the separate flocks and the pooled data.

The general result of this full analysis is that, so far as these six flocks are concerned, the response to dosage as measured by weight is independent of initial egg count; the bad effects of high infestation in any given lamb are sufficiently represented by the low weight with which the lamb enters the experiment. It is therefore reasonable to expect that similar weight improvements would be found in other flocks in which the degree of infestation had resulted in some loss of weight, even if they were less infested than these six, with the helminths that make the main contribution to the total egg-count.

It should be pointed out that the full analysis of Appendix VII very closely confirms the results of Table 4, and just the same estimate of average response, 1.057 lb. per 10 gm., is found as in the above simpler analysis. From the present data the same result could have been obtained, with about the same precision, if initial egg counts had been ignored as to their effects on final weight. But the full analysis was necessary to bring out this fact, and may well be necessary with any subsequent body of data.

The analyses of Table 4 and Appendix VII take account of the initial and final weights of the outside flocks. But there were actually six weighings, in all, as shown in Appendix IV, and it may be of some interest to present these in graphical form, pooling the data of the six flocks. Figure 13 shows a separate curve for each level of dosage and the curves are made comparable by using as a vertical scale the average gain in weight over the initial weight; the horizontal scale is the time in weeks. The curves agree in reaching a maximum between the sixth and twelfth weeks and then falling off. This might suggest that the good effects of dosing were wearing off by about the ninth week and that re-infestation was responsible for the decline. But it should be observed that the curve for the untreated controls is of the same shape as the rest, and it seems likely that the decreasing weight is merely a seasonal effect; at most centres the ninth week fell in December when loss of weight in lambs is a common occurrence.

It is true that the falls in weight for the controls and for the lambs receiving 5 or 10 gm. of phenothiazine start somewhat earlier, and are proportionately rather greater than those for the groups receiving 20, 40 or 50 gm. of phenothiazine. But these differences might well be due to other factors, and no indications are given by this series of trials as to what might have occurred if a second dose of phenothiazine had been administered at some interval after the first.

The inside flocks do not afford satisfactory material for assessing the effect of phenothiazine, at the various dose levels employed, on gain in weight, since the final weighings were made only five weeks after the administration of the drug. For comparative purposes, however, the relevant figures, calculated in the same way as for the outside flocks, are set out in Table 5.

TABLE 5

INCREASES IN LIVE WEIGHT COMPARED WITH UNTREATED LAMBS, IN LB. PER LAMB. INSIDE FLOCKS

| *Dosage in gm.* | *Weybridge* | *Cambridge* | *St. Albans* | *Edinburgh* | *Average* |
|---|---|---|---|---|---|
| 5 | —2.40 | —1.40 | 0.97 | 2.95 | 0.13 |
| 10 | 2.81 | —1.37 | 3.97 | 2.32 | 1.93 |
| 20 | 0.96 | —1.56 | 11.81 | 1.63 | 3.21 |
| 30 | 1.65 | —4.61 | 2.88 | 3.16 | 0.78 |
| 40 | —0.03 | —4.92 | 7.90 | 3.62 | 1.64 |
| 50 | 3.64 | —3.40 | —0.08 | 0.23 | 0.10 |

It will be seen that, beyond the fact that the averages for all treated groups show an advantage over the untreated, the figures yield no significant information. The high average value for the 20 gm. dose depends on the unusually high figure recorded for this dose at a single centre.

This result is not surprising. Reference to Figure 13 shows that, three weeks after dosage, the gains in weight of the different outside groups showed no relation to dosage level. This relation is beginning to be established by the sixth week, and is obvious by the ninth. The two sets of observations are therefore confirmatory.

(d) *The Effect of Drenches as Compared with Tablets*

One of the comparisons which the trials were designed to make was that between two methods of dispensing the drug: (i) suspended in water and given as a drench, and (ii) in the form of compressed tablets given as such. With this end in view drenches were made up so as to contain the same net weight of phenothiazine as the appropriate whole number of tablets given at each dosage level, and the same number of animals received the drug in each of the two forms. On the question of relative efficacy it can be said at once that there is very little evidence for a decision either way, and what evidence can be found is conflicting.

Thus, the " dosage-mortality " curve, applied to the inside flock's egg counts at Edinburgh (Appendix VI) showed that tablets were generally more effective in reducing the egg count than drenches (except at the 10 gm. dosage where drenches were superior to tablets, and both gave egg counts lower than expected). It is thought that this result was due to chance since it could not be confirmed at other centres.

The charts of worm counts, based on pooled data from the four centres, reveal mainly random differences between tablets and drenches, the respective curves being interlaced in the case of most worms. But it is at least suggestive that the drenches curve is consistently lower than the tablets curve for the abomasal worms *Haemonchus, Ostertagia,* and *Trichostrongylus axei.*

Unpublished observations made at St. Albans had indicated that drenches containing as little as 0.15 gm. and 0.3 gm. phenothiazine per Kg. live weight had greatly reduced the count of *Haemonchus* worms in lambs when wetting agents were added to the drenches, whereas there had been no reduction at all when the same amounts of drug were given without any wetting agent. Phenothiazine is rarely given in practice without wetting agents, except for red-worms in horses, where it appears to be fully effective. This suggests the possibility that the presence of wetting agents and their nature are important factors up to the point where the drug reaches the duodenum, when the host's bile salts would presumably function themselves as wetting agents.

The present trials were not designed to test the effects of wetting agents but, in the light of the previous suggestion, it may well be that the apparent superiority of drenches, in counts of worms from the fourth stomach only, is a real effect. Little is known about

the exact behaviour of freshly made tablets after they are swallowed, but if the minute particles of phenothiazine in a tablet are not fully dispersed until the drug comes into contact with bile in the duodenum, the suggestive anomaly in the abomasal worm counts would be fully explained.

## 5.—Summary

The observations recorded in this series of experiments afford data on the effect of phenothiazine, administered as a single dose at varying dose levels, on (*a*) the total egg count, (*b*) the count of different species of helminths in different regions of the intestinal tract, and (*c*) the growth curve of the lambs as judged by changes in weight. Information in regard to (*a*) was obtained mainly from the inside flocks, in regard to (*b*) solely from the inside flocks, and in regard to (*c*) mainly from the outside flocks.

The recorded results emphasize the advantage in investigations of this kind of planned trials carried out by the same technique at several co-operating centres. The uncontrollable variables are so many that differences in results are always to be expected. Reference to the Tables in the text, or in the Appendices, shows clearly what misleading conclusions might have been drawn from the results recorded from any one centre. It is equally clear that quantitative results of this kind require, for their correct appreciation, an adequate statistical analysis, and that such an analysis is only possible when the trials are planned with this point in view. Statistical analysis cannot be satisfactorily applied to a heterogeneous mass of recorded data collected with no uniformity of plan, and without reference to the factors which make it possible to obtain the maximal amount of information from an experiment planned on any given scale.

When we compare the findings obtained under the various headings set out above, several points of interest emerge. The main fall in total egg count was obtained, in the inside flocks, with the first 5 gm. of phenothiazine administered, and this is confirmed in the two outside flocks in which the initial effect of the phenothiazine was not obscured by subsequent re-infestation.

The worm counts indicate that the stomach worms, which contribute largely to the egg counts in the fæces, are greatly reduced at the lower levels of phenothiazine dosage, while the intestinal worms require larger doses to effect any significant reduction. Some intestinal helminths show higher counts at the higher dose levels employed, and this is particularly the case with *Nematodirus,* which contributes in only a minor degree to the total egg count.

Turning to the data in regard to gain in weight of the lambs, derived from the outside flocks, we find a progressive advantage with increasing dosage. The smaller doses, 5 and 10 gm., though they produced the most striking fall in the fæcal egg count, and a marked fall in the stomach worm count, had no effect in promoting gain in weight. Indeed, at all weighings from the ninth week onwards, these groups did worse than the controls, and the disadvantage is particularly marked in the lambs receiving the 10 gm. dose, as shown by the crude data.

Increasing the dose of phenothiazine from 10 to 20 gm. had little effect on the total fæcal egg count, or on the worm count of *Haemonchus;* but it produced a considerable further fall in the worm count of *Trichostrongylus axei* and of *Ostertagia.* It also led to a significant gain in weight as compared with the control group, or the groups receiving 5 or 10 gm. of phenothiazine.

Increases in dose level beyond 20 gm. had little effect either on the total fæcal egg count, or on the count of stomach worms, but they reduced the counts of the intestinal worms, *Bunostomum, Trichostrongylus* spp. and *Cooperia,* and resulted in progressive increases in weight. Whether this indicates that these intestinal worms have a more adverse effect on the health of lambs than has commonly been supposed is a question that merits further investigation.

## 6.—Conclusions

1. Phenothiazine has been tested in single doses against sheep nematodes in co-ordinated trials involving some 280 lambs at a number of centres in Great Britain, using lamb weights, egg counts, and worm counts as criteria of efficacy. Full details are given of the methods used, and it is shown that egg and worm counting techniques are satisfactory.

2. The unit flock, omitting reserves, consisted of 28 lambs given the following doses of phenothiazine in batches of four; 0 (controls), 5 gm., 10 gm., 20 gm., 30 gm., 40 gm., and 50 gm. In each batch except the controls, two lambs received the drug as compressed tablets and two as powder made into a drench with water. About 2 per cent. sodium cetyl sulphate was used as a wetting and dispersing agent in both cases.

3. Four of these unit flocks, maintained inside for six weeks to avoid re-infestation, were tested respectively at Cambridge, Weybridge, St. Albans, and Edinburgh; they supplied information mainly on egg and worm counts.

4. Six unit flocks kept outside for sixteen weeks under natural conditions and tested at St. Albans, Edinburgh, and four commercial farms near Weybridge, supplied information mainly on the net effects of treatment on growth of lambs.

5. The evidence of the inside flocks indicated that the greatest relative reduction in egg count was achieved by the lowest dose used (5 gm.), higher doses reducing the count further to a relatively small extent and irregularly. From the worm counts it appeared that the marked susceptibility of *Haemonchus* to a 5 gm. dose was largely responsible for this effect. With most intestinal worms the response was less marked but continued up to the highest dose used. Anomalous responses were obtained from *Strongyloides, Trichuris, Nematodirus,* and *Moniezia,* the last two actually showing *increased* counts with higher doses.

In an experiment lasting six weeks, with the lambs kept under conditions precluding re-infestation, an actual increase in any species is practically impossible. If these apparent increases are judged significant, the reason must be that the normal elimination of these worms is reduced.

6. The evidence of the outside flocks, maintained for sixteen weeks, was that, while egg count reductions had been obliterated by re-infestation (except in flocks grazed on unusually clean pastures), there was a relatively large weight response which increased roughly in proportion to dosage up to the highest dose used. This response averaged 48 times the weight of phenothiazine administered. The general impression here, viz., that the higher the dose (within the range explored), the better the result, indicated that, judging from the " inside " worm counts, this result must be ascribed to the reduction not merely of stomach worms but also of certain intestinal worms usually considered of little clinical importance.

7. A detailed analysis of the data relating to the outside flocks showed that, within the six flocks tested, the weight responses were not affected by the initial degree of infestation (as measured by egg counts) with which each lamb entered the experiment. There is thus no confirmation of the suggestion that heavily infested lambs respond less well than lightly infested lambs. If it can be assumed that these lambs were not greatly above the average level of infestation for lambs of the same age in this country, then it is likely that the average flock will respond at least as well as these.

8. There is no evidence of any difference in efficacy between tablets and drenches containing the same weight of phenothiazine, beyond a suggestion from the worm counts (which are too variable for exact statistical treatment) that drenches may be slightly more efficacious against stomach worms only. If so, the explanation may be that the particles of phenothiazine in a tablet are not fully dispersed until they are mixed with the host's bile salts.

9. This Report contains two new developments of existing statistical procedures: (i) the method for transforming the sigmoid " dosage mortality " curve to a straight line is extended to cover data like egg counts, where expectations are calculable from prior observations (Appendix VI). (ii) The method of analyzing multiple regressions by the use of the " covariance matrix " is developed so as to cope with a series of samples which are pooled with respect to some variates but treated independently with respect to others (Appendix VII).

We would wish to express our thanks to H. E. Bowman, W. Buist, and G. C. Martin for their technical assistance.

## APPENDICES

### APPENDIX I. Time-table of Experiments

| | OUTSIDE FLOCK | | | INSIDE FLOCK | | |
|---|---|---|---|---|---|---|
| Week | Monday | Wednesday | Friday | Monday | Wednesday | Friday |
| 1 | 1st | 2nd | 3rd Egg count<br>1st Weighing<br>Dosing | Acclimatization | | |
| 2 | | | | 1st | 2nd | 3rd Egg count<br>1st Weighing: Dosing |
| 4 | | | 2nd Weighing | | | |
| 5 | | | | 4th | 5th | 6th Egg count |
| 6 | | | | 7th | 8th | 9th Egg count |
| 7 | | | 3rd Weighing | 2nd Weighing and Killing (followed by worm counting at convenience) | | |
| 10 | | | 4th Weighing | | | |
| 13 | | | 5th Weighing | | | |
| 15 | 4th | 5th | 6th Egg count | | | |
| 16 | 7th | 8th | 9th Egg count<br>6th Weighing | | | |

# APPENDIX II

## Inside Flocks : Initial and Final Total Egg Counts and Lamb Weights

(Initial egg counts, sums of three counts on separate days, have been doubled to make them comparable with final egg counts, which are sums of six counts. For statistical computation the tabulated initial counts should therefore be halved.)

| Dose in gm. | Administration | EDINBURGH | | | | ST. ALBANS | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | TOTAL EGG COUNTS | | WEIGHTS (in lb.) | | TOTAL EGG COUNTS | | WEIGHTS (in. lb.) | |
| | | Initial × 2 | Final × 1 | Initial | Final | Initial × 2 | Final × 1 | Initial | Final |
| 0 | T* | 202 | 175 | 48 | 51 | 190 | 253 | 66.00 | 71.00 |
| | | 182 | 126 | 41 | 43 | 676 | 594 | 54.50 | 56.00 |
| | D* | 150 | 184 | 37 | 36 | 530 | 567 | 57.25 | 55.50 |
| | | 108 | 135 | 36 | 36 | 842 | 1,769 | 62.00 | 58.25 |
| | | 642 | 620 | 162 | 166 | 2,238 | 3,183 | 239.75 | 240.75 |
| 5 | T | 232 | 38 | 34 | 39 | 1,314 | 376 | 73.75 | 75.75 |
| | | 776 | 136 | 47 | 47 | 622 | 286 | 91.00 | 92.50 |
| | D | 224 | 66 | 27 | 26 | 150 | 137 | 60.25 | 60.75 |
| | | 154 | 95 | 38 | 45 | 670 | 784 | 75.25 | 70.75 |
| | | 1,386 | 335 | 146 | 157 | 2,756 | 1,583 | 300.25 | 299.75 |
| 10 | T | 262 | 47 | 31 | 32 | 404 | 294 | 51.25 | 60.00 |
| | | 286 | 43 | 41 | 42 | 442 | 378 | 52.25 | 52.25 |
| | D | 506 | 41 | 28 | 29 | 386 | 473 | 62.25 | 68.00 |
| | | 248 | 37 | 34 | 38 | 232 | 148 | 78.75 | 87.75 |
| | | 1,302 | 168 | 134 | 141 | 1,464 | 1,293 | 244.50 | 268.00 |
| 20 | T | 126 | 21 | 35 | 38 | 1,632 | 107 | 38.25 | 43.50 |
| | | 160 | 17 | 33 | 34 | 1,000 | 79 | 49.50 | 61.00 |
| | D | † — | — | — | — | 250 | 49 | 57.75 | 72.50 |
| | | 208 | 41 | 34 | 35 | 308 | 166 | 61.00 | 70.00 |
| | | 494 | 79 | 102 | 107 | 3,190 | 401 | 206.50 | 247.00 |
| 30 | T | 108 | 21 | 41 | 44 | 170 | 143 | 65.00 | 73.50 |
| | | 574 | 31 | 34 | 39 | 326 | 217 | 74.25 | 71.25 |
| | D | 444 | 55 | 37 | 40 | 256 | 81 | 66.25 | 70.00 |
| | | 122 | 47 | 39 | 41 | 662 | 252 | 78.75 | 88.50 |
| | | 1,248 | 154 | 151 | 164 | 1,414 | 693 | 284.25 | 303.25 |
| 40 | T | 182 | 2 | 44 | 46 | 660 | 154 | 45.25 | 50.25 |
| | | 212 | 18 | 41 | 47 | 394 | 126 | 47.75 | 53.25 |
| | D | 248 | 39 | 41 | 48 | 1,436 | 114 | 50.25 | 54.75 |
| | | 92 | 2 | 35 | 38 | 222 | 87 | 77.50 | 91.25 |
| | | 734 | 61 | 161 | 179 | 2,712 | 481 | 220.75 | 249.50 |
| 50 | T | 224 | 4 | 42 | 42 | 282 | 56 | 71.00 | 77.25 |
| | | 122 | 5 | 41 | 43 | 570 | 129 | 58.25 | 53.00 |
| | D | 92 | 37 | 42 | 44 | 606 | 647 | 70.00 | 67.50 |
| | | 96 | 3 | 42 | 44 | 180 | 92 | 64.75 | 71.75 |
| | | 534 | 49 | 167 | 173 | 1,638 | 924 | 264.00 | 269.50 |

* T = Tablets, D = Drench, † died.

INSIDE FLOCKS : INITIAL AND FINAL TOTAL EGG COUNTS AND LAMB WEIGHTS

(Initial egg counts, sums of three counts on separate days, have been doubled to make them comparable with final egg counts, which are sums of six counts. For statistical computation the tabulated initial counts should therefore be halved.)

| Dose in gm. | Administration | WEYBRIDGE | | | | CAMBRIDGE | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | TOTAL EGG COUNTS | | WEIGHTS (in lb.) | | TOTAL EGG COUNTS | | WEIGHTS (in lb.) | |
| | | Initial × 2 | Final × 1 | Initial | Final | Initial × 2 | Final × 1 | Initial | Final |
| 0 | T* | 250 | 212 | 76.5 | 79.0 | 126 | 102 | 64.75 | 57.75 |
| | D* | 242 | 291 | 96.0 | 96.5 | 358 | 364 | 63.50 | 54.50 |
| | | † — | — | — | — | 608 | 510 | 72.50 | 74.75 |
| | | 192 | 76 | 86.0 | 91.5 | 150 | 165 | 93.25 | 88.50 |
| | | 684 | 579 | 258.5 | 267.0 | 1,242 | 1,141 | 294.00 | 275.50 |
| 5 | T | 308 | 61 | 94.0 | 95.0 | 1,146 | 919 | 57.25 | 52.00 |
| | D | 260 | 119 | 69.5 | 69.0 | 560 | 236 | 88.75 | 80.50 |
| | | 122 | 61 | 71.0 | 70.0 | 530 | 100 | 87.25 | 80.50 |
| | | 40 | 24 | 86.0 | 90.0 | 300 | 190 | 81.25 | 73.25 |
| | | 730 | 265 | 320.5 | 324.0 | 2,536 | 1,445 | 314.50 | 286.25 |
| 10 | T | 498 | 267 | 68.0 | 80.0 | 214 | 70 | 81.50 | 70.75 |
| | D | 180 | 45 | 77.0 | 85.0 | † — | — | — | — |
| | | 310 | 117 | 89.5 | 87.5 | 770 | 99 | 77.50 | 73.75 |
| | | 292 | 27 | 72.0 | 74.0 | 450 | 54 | 67.25 | 62.50 |
| | | 1,280 | 456 | 306.5 | 326.5 | 1,434 | 223 | 226.25 | 207.00 |
| 20 | T | 2,058 | 2,709 | 54.5 | 40.5 | 172 | 58 | 66.25 | 58.25 |
| | D | 100 | 5 | 71.5 | 74.0 | 368 | 292 | 65.50 | 55.00 |
| | | 122 | 8 | 72.0 | 80.0 | 580 | 102 | 55.00 | 54.75 |
| | | 264 | 70 | 86.0 | 92.0 | 388 | 242 | 92.25 | 88.50 |
| | | 2,544 | 2,792 | 284.0 | 286.5 | 1,508 | 694 | 279.00 | 256.50 |
| 30 | T | 478 | 157 | 92.5 | 95.5 | 444 | 390 | 80.00 | 61.75 |
| | D | 20 | 1 | 89.5 | 95.0 | 406 | 153 | 68.00 | 61.00 |
| | | 374 | 76 | 69.0 | 70.5 | 634 | 45 | 62.00 | 59.00 |
| | | 34 | 6 | 90.0 | 98.0 | 596 | 87 | 68.50 | 61.75 |
| | | 906 | 240 | 341.0 | 359.0 | 2,080 | 675 | 278.50 | 243.50 |
| 40 | T | 208 | 26 | 91.0 | 93.0 | 130 | 340 | 91.75 | 75.75 |
| | D | 222 | 24 | 80.0 | 82.0 | 382 | 589 | 90.50 | 75.50 |
| | | 308 | 34 | 75.5 | 78.0 | 422 | 27 | 74.75 | 70.00 |
| | | 116 | 28 | 72.0 | 77.5 | 608 | 236 | 56.25 | 50.50 |
| | | 854 | 112 | 318.5 | 330.5 | 1,542 | 1,192 | 313.25 | 271.75 |
| 50 | T | 320 | 15 | 88.5 | 99.0 | 314 | 182 | 84.00 | 71.50 |
| | D | 214 | 70 | 85.5 | 86.5 | † — | — | — | — |
| | | 84 | 8 | 70.0 | 80.0 | 218 | 8 | 71.75 | 64.00 |
| | | 166 | 3 | 91.5 | 97.0 | 438 | 251 | 87.25 | 79.75 |
| | | 784 | 96 | 335.5 | 362.5 | 970 | 441 | 243.00 | 215.25 |

* T = Tablets, D = Drench. † died.

# APPENDIX III

## Inside Flocks : Worm Counts : Edinburgh

(Grouped under abomasum, small and large intestine, and then in descending order of average frequency.)

| WORM SPECIES | DOSE IN GM. | | | | | | |
|---|---|---|---|---|---|---|---|
| | 0 | 5 | 10 | 20 | 30 | 40 | 50 |
| ABOMASUM | 780 | 20 | 140 | 0 | 0 | 0 | 0 |
| | 300 | 200 | 120 | 0 | 0 | 0 | 0 |
| 1 *Trichostrongylus axei* | 420 | 0 | 240 | 60 | 0 | 0 | 0 |
| | 360 | 780 | 20 | 20 | 0 | 0 | 0 |
| | 340 | 30 | 60 | 0 | 20 | 0 | 0 |
| 2 *Ostertagia* | 1,150 | 460 | 240 | 60 | 20 | 0 | 0 |
| | 720 | 80 | 300 | 20 | 0 | 0 | 0 |
| | 760 | 900 | 0 | 0 | 0 | 0 | 0 |
| | 24 | 1 | 1 | 0 | 0 | 0 | 0 |
| 3 *Haemonchus* | 22 | 2 | 0 | 0 | 0 | 0 | 0 |
| | 16 | 2 | 2 | 0 | 0 | 0 | 0 |
| | 12 | 3 | 0 | 1 | 0 | 0 | 0 |
| SMALL INTESTINE | 2,220 | 620 | 900 | 520 | 380 | 100 | 140 |
| | 770 | 4,500 | 400 | 140 | 760 | 20 | 20 |
| 4 *Trichostrongylus* spp. | 1,700 | 1,360 | 500 | 640 | 140 | 80 | 120 |
| | 420 | 2,100 | 440 | 700 | 1,020 | 100 | 160 |
| | 1,270 | 720 | 20 | 0 | 620 | 200 | 500 |
| 5 *Nematodirus* | 870 | 1,500 | 1,120 | 440 | 1,600 | 600 | 180 |
| | 5,650 | 320 | 250 | 820 | 1,300 | 2,960 | 460 |
| | 520 | 1,600 | 3,540 | 1,120 | 3,800 | 0 | 260 |
| | 500 | 40 | 400 | 120 | 100 | 20 | 140 |
| 6 *Strongyloides* | 100 | 120 | 420 | 400 | 240 | 140 | 40 |
| | 800 | 120 | 150 | 500 | 660 | 140 | 240 |
| | 260 | 200 | 60 | 160 | 120 | 60 | 80 |
| | 0 | 300 | 0 | 0 | 0 | 40 | 0 |
| 7 *Cooperia* | 50 | 1,900 | 180 | 0 | 0 | 0 | 0 |
| | 500 | 200 | 0 | 60 | 140 | 0 | 60 |
| | 420 | 300 | 140 | 60 | 360 | 0 | 0 |
| | 5 | 16 | 5 | 0 | 0 | 0 | 0 |
| 8 *Bunostomum* | 8 | 19 | 60 | 0 | 0 | 0 | 0 |
| | 14 | 0 | 11 | 0 | 0 | 1 | 0 |
| | 19 | 14 | 1 | 0 | 1 | 0 | 0 |
| | 0 | 3 | 0 | 11 | 0 | 4 | 0 |
| 9 *Capillaria* | 4 | 0 | 0 | 3 | 0 | 3 | 4 |
| | 0 | 0 | 11 | 0 | 0 | 3 | 2 |
| | 0 | 1 | 0 | 0 | 5 | 0 | 0 |
| | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 10 *Moniezia* | 0 | 0 | 1 | 0 | 0 | 5 | 0 |
| | 1 | 3 | 0 | 0 | 0 | 1 | 0 |
| | 0 | 0 | 0 | 0 | 0 | 1 | 0 |
| LARGE INTESTINE | 35 | 13 | 19 | 8 | 14 | 10 | 9 |
| | 12 | 7 | 28 | 9 | 13 | 10 | 0 |
| 11 *Trichuris* | 4 | 5 | 25 | 11 | 12 | 6 | 15 |
| | 2 | 8 | 12 | 7 | 3 | 19 | 15 |
| | 0 | 2 | 3 | 0 | 2 | 0 | 0 |
| 12 *Oesophagostomum* | 22 | 6 | 3 | 0 | 0 | 0 | 0 |
| | 1 | 0 | 0 | 0 | 0 | 1 | 0 |
| | 0 | 0 | 0 | 0 | 1 | 0 | 0 |
| | 2 | 1 | 0 | 0 | 0 | 0 | 0 |
| 13 *Chabertia* | 3 | 2 | 0 | 0 | 0 | 0 | 0 |
| | 2 | 0 | 0 | 0 | 0 | 0 | 0 |
| | 0 | 1 | 0 | 0 | 0 | 0 | 0 |

## APPENDIX III—*continued*

### Inside Flocks : Worm Counts : St. Albans

(Grouped under abomasum, small and large intestine, and then in descending order of average frequency.)

| WORM SPECIES | DOSE IN GM. | | | | | | |
|---|---|---|---|---|---|---|---|
| | 0 | 5 | 10 | 20 | 30 | 40 | 50 |
| ABOMASUM | 2,075 | 8,500 | 550 | 0 | 0 | 0 | 0 |
| | 4,050 | 3,500 | 1,750 | 325 | 0 | 0 | 0 |
| 1 *Trichostrongylus axei* | 7,300 | 100 | 2,600 | 0 | 0 | 50 | 0 |
| | 6,500 | 1,650 | 300 | 25 | 75 | 50 | 0 |
| | 800 | 7,900 | 900 | 50 | 100 | 425 | 0 |
| 2 *Ostertagia* | 1,650 | 2,700 | 900 | 225 | 25 | 0 | 25 |
| | 2,600 | 500 | 1,550 | 0 | 0 | 50 | 0 |
| | 6,500 | 3,250 | 525 | 0 | 0 | 25 | 50 |
| | 289 | 3 | 0 | 0 | 0 | 0 | 0 |
| 3 *Haemonchus* | 378 | 81 | 0 | 0 | 0 | 0 | 0 |
| | 415 | 4 | 2 | 0 | 0 | 1 | 0 |
| | 682 | 1 | 1 | 0 | 0 | 0 | 0 |
| SMALL INTESTINE | 12,000 | 1,250 | 27,000 | 6,050 | 11,200 | 925 | 200 |
| | 21,000 | 21,200 | 32,600 | 850 | 3,400 | 867 | 250 |
| 4 *Trichostrongylus* spp. | 11,600 | 4,500 | 24,100 | 36,000 | 900 | 638 | 1,150 |
| | 28,800 | 31,400 | 6,150 | 2,033 | 10,000 | 1,533 | 225 |
| | 0 | 650 | 0 | 0 | 2,600 | 600 | 3,275 |
| 5 *Nematodirus* | 0 | 200 | 200 | 250 | 2,600 | 3,267 | 1,225 |
| | 0 | 450 | 0 | 0 | 3,133 | 150 | 1,700 |
| | 300 | 300 | 500 | 0 | 1,500 | 633 | 650 |
| | 200 | 600 | 500 | 100 | 900 | 550 | 400 |
| 6 *Strongyloides* | 200 | 600 | 200 | 400 | 800 | 1,100 | 450 |
| | 0 | 250 | 1,400 | 1,000 | 500 | 638 | 2,700 |
| | 600 | 600 | 1,350 | 1,100 | 1,600 | 733 | 750 |
| | 400 | 50 | 2,200 | 100 | 500 | 0 | 25 |
| 7 *Cooperia* | 0 | 200 | 2,600 | 550 | 100 | 33 | 13 |
| | 300 | 50 | 0 | 2,600 | 33 | 13 | 0 |
| | 200 | 900 | 200 | 67 | 100 | 0 | 0 |
| | 4 | 19 | 3 | 3 | 1 | 0 | 0 |
| 8 *Bunostomum* | 1 | 11 | 1 | 0 | 1 | 0 | 0 |
| | 3 | 4 | 6 | 0 | 0 | 0 | 0 |
| | 18 | 3 | 1 | 1 | 0 | 0 | 0 |
| | 0 | 0 | 0 | 0 | 0 | 2 | 9 |
| 10 *Moniezia* | 0 | 1 | 0 | 8 | 7 | 0 | 3 |
| | 0 | 2 | 0 | 0 | 0 | 3 | 21 |
| | 3 | 0 | 4 | 4 | 1 | 2 | 1 |
| LARGE INTESTINE | 3 | 18 | 4 | 26 | 11 | 44 | 4 |
| | 1 | 18 | 4 | 4 | 10 | 15 | 20 |
| 11 *Trichuris* | 2 | 47 | 2 | 2 | 13 | 66 | 18 |
| | 57 | 59 | 8 | 0 | 5 | 5 | 0 |
| | 0 | 4 | 0 | 0 | 0 | 0 | 0 |
| 12 *Oesophagostomum* | 33 | 3 | 8 | 0 | 0 | 1 | 0 |
| | 10 | 1 | 0 | 0 | 6 | 0 | 1 |
| | 0 | 10 | 0 | 15 | 10 | 0 | 0 |
| | 30 | 25 | 9 | 4 | 7 | 22 | 2 |
| 13 *Chabertia* | 0 | 137 | 40 | 3 | 4 | 17 | 0 |
| | 26 | 0 | 1 | 4 | 10 | 1 | 4 |
| | 132 | 3 | 5 | 16 | 3 | 1 | 2 |

## APPENDIX III—*continued*

### Inside Flocks : Worm Counts : Weybridge

(Grouped under abomasum, small and large intestine, and then in descending order of average frequency.)

| | Worm species | Dose in gm. | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | 0 | 5 | 10 | 20 | 30 | 40 | 50 |
| Abomasum | | 2,125 | 5,650 | 3,725 | 1,025 | 1,700 | 50 | 50 |
| | | 5,313 | 2,100 | 2,400 | 0 | 0 | 200 | 550 |
| 1 | *Trichostrongylus axei* | 8,700* | 1,375 | 3,225 | 25 | 200 | 350 | 0 |
| | | 9,150 | 100 | 3,250 | 0 | 50 | 25 | 0 |
| | | 925 | 550 | 400 | 850 | 250 | 125 | 25 |
| 2 | *Ostertagia* | 891 | 1,350 | 850 | 50 | 0 | 125 | 300 |
| | | 500* | 1,175 | 225 | 25 | 300 | 25 | 25 |
| | | 900 | 175 | 400 | 25 | 0 | 50 | 75 |
| | | 275 | 15 | 1 | 8 | 4 | 0 | 0 |
| 3 | *Haemonchus* | 363 | 25 | 5 | 0 | 0 | 0 | 0 |
| | | 0* | 0 | 0 | 0 | 5 | 0 | 0 |
| | | 93 | 0 | 0 | 0 | 0 | 2 | 0 |
| Small intestine | | 1,975 | 4,666 | 4,900 | 11,200 | 6,950 | 2,550 | 800 |
| | | 4,200 | 125 | 975 | 350 | 275 | 1,050 | 3,225 |
| 4 | *Trichostrongylus* spp. | 6,150* | 625 | 2,575 | 300 | 3,840 | 1,850 | 275 |
| | | 675 | 650 | 300 | 4,719 | 250 | 1,250 | 575 |
| | | 125 | 0 | 750 | 0 | 100 | 550 | 350 |
| 5 | *Nematodirus* | 233 | 100 | 500 | 150 | 800 | 50 | 700 |
| | | 500* | 25 | 25 | 50 | 690 | 200 | 1,100 |
| | | 0 | 250 | 0 | 66 | 175 | 475 | 275 |
| | | 775 | 1,133 | 1,100 | 54,400 | 1,200 | 1,125 | 0 |
| 6 | *Strongyloides* | 1,466 | 600 | 1,025 | 75 | 150 | 150 | 275 |
| | | 150* | 475 | 400 | 500 | 1,680 | 175 | 225 |
| | | 575 | 150 | 850 | 858 | 650 | 250 | 0 |
| | | 1,875 | 1,000 | 4,750 | 6,000 | 3,150 | 250 | 950 |
| 7 | *Cooperia* | 2,900 | 0 | 275 | 175 | 50 | 550 | 950 |
| | | 5,250* | 275 | 1,250 | 0 | 420 | 25 | 0 |
| | | 0 | 125 | 150 | 1,221 | 0 | 50 | 50 |
| | | 155 | 94 | 26 | 200 | 0 | 3 | 26 |
| 8 | *Bunostomum* | 66 | 45 | 40 | 0 | 1 | 0 | 38 |
| | | 35* | 43 | 193 | 10 | 12 | 7 | 0 |
| | | 41 | 0 | 23 | 54 | 0 | 0 | 1 |
| | | 25 | 0 | 0 | 0 | 0 | 0 | 0 |
| 9 | *Capillaria* | 0 | 25 | 25 | 25 | 25 | 0 | 0 |
| | | 0* | 0 | 0 | 0 | 0 | 0 | 25 |
| | | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| | | 0 | 0 | 0 | 1 | 0 | 2 | 0 |
| 10 | *Moniezia* | 0 | 0 | 0 | 0 | 0 | 0 | 3 |
| | | 0 | 0 | 0 | 0 | 1 | 1 | 1 |
| | | 1 | 0 | 0 | 3 | 0 | 0 | 1 |
| Large intestine | | 0 | 6 | 86 | 348 | 32 | 19 | 21 |
| | | 41 | 1 | 95 | 1 | 1 | 12 | 0 |
| 11 | *Trichuris* | 128* | 30 | 11 | 11 | 23 | 21 | 9 |
| | | 1 | 0 | 12 | 1 | 0 | 17 | 5 |
| | | 1 | 1 | 5 | 0 | 0 | 0 | 1 |
| 12 | *Oesophagostomum* | 3 | 3 | 18 | 0 | 0 | 2 | 0 |
| | | 0* | 16 | 1 | 18 | 1 | 0 | 0 |
| | | 11 | 0 | 0 | 5 | 0 | 0 | 0 |
| | | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 13 | *Chabertia* | 0 | 0 | 0 | 0 | 1 | 0 | 0 |
| | | 0* | 0 | 0 | 0 | 1 | 1 | 0 |
| | | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

* This sheep died 28/12/41.

## Inside Flocks : Worm Counts : Cambridge

(Grouped under abomasum, small and large intestine, and then in descending order of average frequency.)

| Worm species | Dose in gm. | | | | | | |
|---|---|---|---|---|---|---|---|
| | 0 | 5 | 10 | 20 | 30 | 40 | 50 |
| ABOMASUM | | | | | | | |
| | 0 | 2,200 | 975 | 25 | 725 | 0 | 75 |
| | 2,450 | 0 | 4,675 | 0 | 0 | 14,600 | * |
| 1 *Trichostrongylus axei* | 1,250 | 500 | 0 | 0 | 0 | 0 | 0 |
| | 200 | 450 | 0 | 0 | 0 | 0 | 0 |
| | 325 | 2,250 | 1,125 | 100 | 850 | 25 | 50 |
| 2 *Ostertagia* | 1,125 | 0 | 350 | 125 | 0 | 18,300 | * |
| | 1,425 | 525 | 525 | 0 | 75 | 50 | 0 |
| | 50 | 725 | 550 | 150 | 25 | 0 | 0 |
| | 125 | 0 | 25 | 0 | 0 | 0 | 25 |
| 3 *Haemonchus* | 125 | 0 | 0 | 0 | 0 | 0 | * |
| | 350 | 50 | 0 | 0 | 0 | 0 | 0 |
| | 100 | 25 | 0 | 0 | 0 | 0 | 0 |
| SMALL INTESTINE | | | | | | | |
| | 0 | 2,225 | 2,675 | 1,175 | 9,000 | 2,900 | 2,000 |
| | 5,700 | 0 | 33,400 | 250 | 775 | 6,233 | * |
| 4 *Trichostrongylus* spp. | 2,100 | 2,350 | 0 | 1,300 | 1,025 | 50 | 275 |
| | 1,400 | 5,100 | 2,325 | 3,367 | 0 | 275 | 2,500 |
| | 0 | 50 | 0 | 0 | 0 | 0 | 0 |
| 5 *Nematodirus* | 33 | 0 | 200 | 25 | 25 | 0 | * |
| | 0 | 0 | 0 | 175 | 0 | 75 | 50 |
| | 525 | 167 | 0 | 900 | 25 | 100 | 0 |
| | 0 | 975 | 325 | 550 | 1,450 | 650 | 600 |
| 6 *Strongyloides* | 767 | 0 | 0 | 375 | 750 | 533 | * |
| | 50 | 450 | 0 | 450 | 375 | 50 | 100 |
| | 75 | 267 | 175 | 600 | 450 | 1,000 | 1,750 |
| | 0 | 325 | 0 | 0 | 1,350 | 100 | 75 |
| 7 *Cooperia* | 0 | 25 | 100 | 0 | 575 | 0 | * |
| | 3,125 | 1,325 | 0 | 550 | 250 | 0 | 0 |
| | 250 | 2,500 | 425 | 1,800 | 0 | 275 | 75 |
| | 0 | 150 | 50 | 50 | 0 | 0 | 50 |
| 8 *Bunostomum* | 0 | 0 | 100 | 25 | 200 | 33 | * |
| | 50 | 50 | 0 | 0 | 0 | 0 | 0 |
| | 0 | 33 | 0 | 0 | 25 | 0 | 0 |
| LARGE INTESTINE | 375 | 50 | 50 | 75 | 275 | 50 | 75 |
| | 100 | 100 | 2,175 | 850 | 275 | 1,450 | * |
| 11 *Trichuris* | 350 | 175 | 300 | 50 | 25 | 50 | 175 |
| | 25 | 200 | 0 | 200 | 50 | 325 | 50 |
| | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 12 *Oesophagostomum* | 0 | 0 | 50 | 0 | 50 | 0 | * |
| | 200 | 25 | 125 | 0 | 0 | 0 | 0 |
| | 50 | 50 | 25 | 0 | 0 | 0 | 0 |
| | 0 | 400 | 0 | 50 | 0 | 0 | 0 |
| 13 *Chabertia* | 0 | 0 | 25 | 0 | 50 | 75 | * |
| | 550 | 25 | 0 | 0 | 0 | 0 | 0 |
| | 25 | 525 | 50 | 0 | 0 | 0 | 0 |

* This sheep died 4/12/41.

# APPENDIX IV

## Outside Flocks : Initial and Final Total Egg Counts, and Lamb Weights at 3-weekly intervals

(Initial egg counts, sums of three counts on separate days, have been doubled to make them comparable with final egg counts, which are sums of six counts. For statistical computation the tabulated initial counts should therefore be halved.)

EDINBURGH

| Dose in Gm. | Administration | TOTAL EGG COUNTS | | WEIGHTS IN LB. AT: | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Initial ×2 | Final ×1 | 1st week | 4th week | 7th week | 10th week | 13th week | 16th week |
| 0 | T* | 88 | 28 | 37 | 43 | 48 | 50 | 47 | 45 |
| | | 468 | 672 | 27 | 28 | 31 | 32 | 31 | 27 |
| | D* | 132 | 70 | 40 | 43 | 49 | 51 | 52 | 52 |
| | | 176 | 48 | 52 | 54 | 58 | 59 | 59 | 56 |
| | T | 76 | 95 | 42 | 44 | 47 | 48 | 48 | 44 |
| | | 232 | 209 | 57 | 60 | 63 | 64 | 62 | 60 |
| | D | 384 | 305 | 37 | 47 | 53 | 56 | 54 | 52 |
| | | 412 | 131 | 34 | 38 | 43 | 45 | 44 | 41 |
| | | 1,968 | 1,558 | 326 | 357 | 392 | 405 | 397 | 377 |
| 5 | T | † — | † — | — | — | — | — | — | — |
| | | 236 | 84 | 36 | 41 | 45 | 45 | 43 | 41 |
| | D | 378 | 124 | 38 | 40 | 47 | 48 | 44 | 43 |
| | | 72 | 38 | 45 | 47 | 51 | 54 | 54 | 52 |
| | | 686 | 246 | 119 | 128 | 143 | 147 | 141 | 136 |
| 10 | T | 82 | 162 | 40 | 43 | 44 | 45 | 41 | 37 |
| | | 186 | 56 | 44 | 49 | 53 | 53 | 51 | 48 |
| | D | 64 | 22 | 44 | 48 | 51 | 53 | 52 | 53 |
| | | 492 | 109 | 55 | 58 | 58 | 58 | 54 | 48 |
| | | 824 | 349 | 183 | 198 | 206 | 209 | 198 | 186 |
| 20 | T | 160 | 44 | 40 | 43 | 47 | 49 | 48 | 44 |
| | | 362 | 71 | 51 | 54 | 57 | 60 | 60 | 55 |
| | D | 174 | 95 | 38 | 42 | 47 | 48 | 50 | 46 |
| | | 278 | 83 | 37 | 36 | 43 | 47 | 46 | 45 |
| | | 974 | 293 | 166 | 175 | 194 | 204 | 204 | 190 |
| 30 | T | 152 | 67 | 36 | 39 | 44 | 47 | 47 | 43 |
| | | 142 | 50 | 47 | 52 | 56 | 58 | 56 | 54 |
| | D | 314 | 119 | 34 | 38 | 43 | 48 | 50 | 50 |
| | | 180 | 56 | 46 | 53 | 59 | 59 | 60 | 61 |
| | | 788 | 292 | 163 | 182 | 202 | 212 | 213 | 208 |
| 40 | T | 110 | 41 | 45 | 52 | 53 | 57 | 57 | 59 |
| | | 72 | 52 | 42 | 47 | 51 | 53 | 51 | 47 |
| | D | 60 | 47 | 41 | 44 | 46 | 48 | 47 | 43 |
| | | 204 | 54 | 35 | 38 | 44 | 47 | 47 | 48 |
| | | 446 | 194 | 163 | 181 | 194 | 205 | 202 | 197 |
| 50 | T | 162 | 40 | 44 | 46 | 50 | 52 | 53 | 50 |
| | | 298 | 57 | 30 | 33 | 37 | 39 | 41 | 42 |
| | D | 94 | 57 | 39 | 44 | 52 | 58 | 54 | 53 |
| | | 150 | 10 | 49 | 54 | 57 | 59 | 57 | 56 |
| | | 704 | 164 | 162 | 177 | 196 | 208 | 205 | 201 |

*T = Tablets, D = Drench. † died.

## Outside Flocks : Initial and Final Total Egg Counts, and Lamb Weights at 3-weekly intervals

(Initial egg counts, sums of three counts on separate days, have been doubled to make them comparable with final egg counts, which are sums of six counts. For statistical computation the tabulated initial counts should therefore be halved.)

ST. ALBANS

| Dose in Gm. | Administration | Total egg counts | | Weights in lb. at: | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Initial ×2 | Final ×1 | 1st week | 4th week | 7th week | 10th week | 13th week | 16th week |
| 0 | T* | 346 | 80 | 66.50 | 68.75 | 69.50 | 70.00 | 64.75 | 65.75 |
| | | 112 | 165 | 69.75 | 73.00 | 79.75 | 78.50 | 77.75 | 83.50 |
| | D* | 206 | 124 | 77.00 | 78.25 | 80.75 | 82.25 | 75.00 | 81.00 |
| | | 240 | 148 | 43.25 | 51.50 | 51.75 | 50.50 | 46.00 | 48.75 |
| | | 904 | 517 | 256.50 | 271.50 | 281.75 | 281.25 | 263.50 | 279.00 |
| 5 | T | 210 | 225 | 58.25 | 68.25 | 70.50 | 70.00 | 64.00 | 66.25 |
| | | 482 | 147 | 49.50 | 57.25 | 62.50 | 61.00 | 57.25 | 59.25 |
| | D | 568 | 24 | 79.75 | 82.25 | 86.00 | 88.25 | 84.25 | 84.50 |
| | | 138 | 42 | 86.50 | 88.50 | 93.25 | 97.00 | 87.00 | 90.75 |
| | | 1,398 | 438 | 274.00 | 296.25 | 312.25 | 316.25 | 292.50 | 300.75 |
| 10 | T | 290 | 181 | 67.75 | 74.00 | 74.25 | 70.50 | 67.50 | 66.50 |
| | | 358 | 205 | 71.25 | 72.00 | 79.00 | 77.90 | 73.00 | 70.50 |
| | D | 708 | 237 | 68.25 | 69.75 | 74.75 | 78.50 | 76.25 | 73.25 |
| | | 794 | 207 | 83.25 | 79.25 | 84.75 | 86.50 | 80.25 | 82.50 |
| | | 2,150 | 830 | 290.50 | 295.00 | 312.75 | 312.50 | 297.00 | 292.75 |
| 20 | T | 260 | 118 | 72.50 | 76.75 | 81.50 | 84.75 | 78.00 | 84.25 |
| | | 686 | 159 | 88.25 | 91.50 | 94.75 | 96.50 | 87.50 | 93.25 |
| | D | 506 | 359 | 69.00 | 72.50 | 76.00 | 77.75 | 67.50 | 70.00 |
| | | 486 | 63 | 68.25 | 74.00 | 79.00 | 77.75 | 72.00 | 74.75 |
| | | 1,938 | 699 | 298.00 | 314.75 | 331.25 | 336.75 | 305.00 | 322.25 |
| 30 | T | 876 | 254 | 52.50 | 52.00 | 57.75 | 58.75 | 54.75 | 54.00 |
| | | 174 | 56 | 69.50 | 73.00 | 82.00 | 84.00 | 76.25 | 80.75 |
| | D | 124 | 47 | 61.50 | 69.50 | 75.75 | 79.50 | 74.00 | 74.50 |
| | | 248 | 310 | 87.50 | 89.75 | 95.25 | 95.00 | 87.25 | 84.00 |
| | | 1,422 | 667 | 271.00 | 284.25 | 310.75 | 317.25 | 292.25 | 293.25 |
| 40 | T | 562 | 293 | 68.50 | 73.00 | 76.75 | 72.75 | 76.00 | 79.00 |
| | | 242 | 19 | 63.75 | 71.75 | 73.75 | 80.50 | 75.50 | 78.50 |
| | D | 132 | 143 | 86.25 | 88.75 | 95.00 | 101.75 | 97.50 | 100.75 |
| | | 514 | 193 | 79.75 | 71.75 | 81.50 | 85.25 | 81.00 | 84.75 |
| | | 1,450 | 648 | 298.25 | 305.25 | 327.00 | 340.25 | 330.00 | 343.00 |
| 50 | T | 372 | 87 | 87.00 | 89.75 | 91.25 | 93.50 | 94.50 | 95.25 |
| | | 570 | 284 | 79.00 | 82.75 | 88.00 | 94.00 | 86.50 | 91.75 |
| | D | 278 | 69 | 68.00 | 72.75 | 76.25 | 81.75 | 72.25 | 74.25 |
| | | 336 | 93 | 67.50 | 68.25 | 76.00 | 78.50 | 75.50 | 77.25 |
| | | 1,556 | 533 | 301.50 | 313.50 | 331.50 | 347.75 | 328.75 | 338.50 |

*T = Tablets, D = Drench.

## Outside Flocks : Initial and Final Total Egg Counts, and Lamb Weights at 3-weekly intervals

(Both initial and final egg counts are in this case the sums of three counts on separate days ; initial counts have therefore not been doubled.)

WEYBRIDGE *A*

| Dose in Gm. | Administration | TOTAL EGG COUNTS | | WEIGHTS IN LB. AT: | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Initial ×1 | Final ×1 | 1st week | 4th week | 7th week | 10th week | 13th week | 16th week |
| 0 | T* | 116 | 22 | 42.0 | 49.0 | 53.0 | 57.5 | 56.0 | 56.0 |
| | | † — | — | — | — | — | — | — | — |
| | D* | 137 | 43 | 47.0 | 52.5 | 57.0 | 58.5 | 59.0 | 59.0 |
| | | 54 | 12 | 36.0 | 38.0 | 40.5 | 43.0 | 44.5 | 46.0 |
| | | 307 | 77 | 125.0 | 139.5 | 150.5 | 159.0 | 159.5 | 161.0 |
| 5 | T | 28 | 35 | 48.0 | 53.0 | 57.0 | 60.0 | 59.5 | 59.5 |
| | | † — | — | — | — | — | — | — | — |
| | D | 58 | 9 | 47.0 | 51.0 | 52.0 | 55.5 | 54.5 | 53.5 |
| | | 104 | 81 | 43.0 | 39.5 | 40.0 | 41.0 | 40.0 | 38.5 |
| | | 190 | 125 | 138.0 | 143.5 | 149.0 | 156.5 | 154.0 | 151.5 |
| 10 | T | 51 | 23 | 42.0 | 45.5 | 48.0 | 50.5 | 48.5 | 46.0 |
| | | 17 | 14 | 57.0 | 61.5 | 61.0 | 66.0 | 65.0 | 58.0 |
| | D | 157 | 6 | 35.0 | 42.5 | 43.5 | 49.0 | 50.5 | 47.0 |
| | | 134 | 50 | 44.0 | 50.0 | 57.0 | 62.5 | 61.0 | 57.5 |
| | | 359 | 93 | 178.0 | 199.5 | 209.5 | 228.0 | 225.0 | 208.5 |
| 20 | T | 56 | 13 | 52.0 | 57.0 | 60.0 | 62.0 | 60.0 | 58.0 |
| | | 54 | 23 | 45.0 | 50.0 | 51.0 | 56.0 | 56.0 | 54.0 |
| | D | 116 | 40 | 40.0 | 47.0 | 49.0 | 57.0 | 58.0 | 58.0 |
| | | 54 | 15 | 55.0 | 57.5 | 57.0 | 62.0 | 61.0 | 62.0 |
| | | 280 | 91 | 192.0 | 211.5 | 217.0 | 237.0 | 235.0 | 232.0 |
| 30 | T | 12 | 1 | 55.0 | 60.0 | 66.5 | 71.0 | 70.0 | 69.5 |
| | | † — | — | — | — | — | — | — | — |
| | D | 33 | 15 | 43.0 | 46.0 | 53.0 | 52.0 | 52.0 | 53.0 |
| | | 32 | 13 | 42.0 | 45.0 | 46.0 | 51.5 | 52.5 | 53.0 |
| | | 77 | 29 | 140.0 | 151.0 | 165.5 | 174.5 | 174.5 | 175.5 |
| 40 | T | 82 | 6 | 45.0 | 51.0 | 51.0 | 55.0 | 55.0 | 54.0 |
| | | 97 | 17 | 48.0 | 52.0 | 58.0 | 62.5 | 61.0 | 62.0 |
| | D | † — | — | — | — | — | — | — | — |
| | | 44 | 21 | 45.0 | 50.0 | 54.0 | 57.0 | 58.0 | 59.0 |
| | | 223 | 44 | 138.0 | 153.0 | 163.0 | 174.5 | 174.0 | 175.0 |
| 50 | T | 25 | 47 | 35.0 | 39.0 | 43.0 | 43.0 | 43.0 | 42.5 |
| | | 39 | 20 | 51.0 | 55.0 | 61.0 | 66.0 | 66.0 | 66.0 |
| | D | 95 | 78 | 36.0 | 41.0 | 46.5 | 52.0 | 52.0 | 45.0 |
| | | 48 | 27 | 57.0 | 61.0 | 67.0 | 71.0 | 70.5 | 70.0 |
| | | 207 | 172 | 179.0 | 196.0 | 217.5 | 232.0 | 231.5 | 223.5 |

* T = Tablets, D = Drench. † died.

## Outside Flocks : Initial and Final Total Egg Counts, and Lamb Weights at 3-weekly intervals

(Both initial and final egg counts are in this case the sums of three counts on separate days ; initial counts have therefore not been doubled.)

WEYBRIDGE *B*

| Dose in Gm. | Administration | TOTAL EGG COUNTS | | WEIGHTS IN LB. AT: | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Initial ×1 | Final ×1 | 1st week | 4th week | 7th week | 10th week | 13th week | 16th week |
| 0 | T* | †— | — | — | — | — | — | — | — |
| | | 28 | 61 | 78.0 | 81.5 | 81.0 | 78.0 | 78.0 | 71.0 |
| | D* | 315 | 53 | 73.0 | 79.0 | 75.0 | 76.0 | 78.5 | 73.5 |
| | | 61 | 14 | 79.0 | 81.0 | 82.5 | 82.5 | 81.0 | 78.0 |
| | | 404 | 128 | 230.0 | 241.5 | 238.5 | 236.5 | 237.5 | 222.5 |
| 5 | T | 45 | 55 | 86.0 | 94.0 | 91.5 | 89.0 | 86.5 | 85.0 |
| | | 63 | 29 | 82.0 | 85.0 | 90.0 | 88.0 | 86.0 | 82.5 |
| | D | 27 | 23 | 77.0 | 81.5 | 84.5 | 79.0 | 71.5 | 71.0 |
| | | 85 | 29 | 63.0 | 70.0 | 74.0 | 71.0 | 69.5 | 63.0 |
| | | 220 | 136 | 308.0 | 330.5 | 340.0 | 327.0 | 313.5 | 301.5 |
| 10 | T | 56 | 9 | 84.0 | 86.0 | 79.0 | 78.5 | 78.0 | 77.0 |
| | | 18 | 35 | 58.0 | 60.0 | 60.0 | 61.0 | 57.0 | 56.5 |
| | D | 31 | 32 | 74.0 | 80.5 | 81.0 | 80.0 | 76.0 | 76.0 |
| | | 71 | 35 | 81.0 | 85.0 | 85.0 | 85.5 | 76.0 | 75.5 |
| | | 176 | 111 | 297.0 | 311.5 | 305.0 | 305.0 | 287.0 | 285.0 |
| 20 | T | 26 | 18 | 78.0 | 81.5 | 85.5 | 84.5 | 85.0 | 81.0 |
| | | 14 | 8 | 83.0 | 90.0 | 96.0 | 94.0 | 95.0 | 89.0 |
| | D | 74 | 38 | 81.0 | 82.0 | 81.5 | 79.0 | 77.0 | 75.0 |
| | | 50 | 51 | 78.0 | 88.0 | 90.0 | 92.0 | 95.0 | 88.0 |
| | | 164 | 115 | 320.0 | 341.5 | 353.0 | 349.5 | 352.0 | 333.0 |
| 30 | T | 40 | 50 | 90.0 | 97.0 | 98.5 | 97.0 | 92.0 | 91.0 |
| | | 113 | 203 | 79.0 | 79.0 | 79.0 | 77.0 | 73.0 | 76.0 |
| | D | 42 | 45 | 73.0 | 77.5 | 77.0 | 77.0 | 74.5 | 70.0 |
| | | 34 | 43 | 79.0 | 84.0 | 86.0 | 89.5 | 86.0 | 82.0 |
| | | 229 | 341 | 321.0 | 337.5 | 340.5 | 340.5 | 325.5 | 319.0 |
| 40 | T | 74 | 72 | 76.0 | 84.0 | 87.0 | 88.0 | 84.0 | 81.0 |
| | | †— | — | — | — | — | — | — | — |
| | D | 13 | 11 | 78.0 | 84.0 | 86.5 | 85.0 | 86.0 | 83.0 |
| | | 123 | 15 | 63.0 | 69.0 | 68.0 | 65.0 | 66.0 | 61.0 |
| | | 210 | 98 | 217.0 | 237.0 | 241.5 | 238.0 | 236.0 | 225.0 |
| 50 | T | 101 | 35 | 87.0 | 94.0 | 97.0 | 99.0 | 96.0 | 95.0 |
| | | 28 | 35 | 73.0 | 79.0 | 82.5 | 82.0 | 77.0 | 74.0 |
| | D | 18 | 10 | 74.0 | 80.0 | 80.5 | 79.0 | 77.0 | 76.5 |
| | | 46 | 6 | 86.0 | 89.0 | 93.0 | 91.0 | 89.0 | 86.0 |
| | | 193 | 86 | 320.0 | 342.0 | 353.0 | 351.0 | 339.0 | 331.5 |

* T = Tablets, D = Drench. † died.

OUTSIDE FLOCKS : INITIAL AND FINAL TOTAL EGG COUNTS, AND LAMB WEIGHTS AT 3-WEEKLY INTERVALS

(Both initial and final egg counts are in this case the sums of three counts on separate days ; initial counts have therefore not been doubled.)

WEYBRIDGE *C*

| Dose in Gm. | Administration | TOTAL EGG COUNTS | | WEIGHTS IN LB. AT: | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Initial ×1 | Final ×1 | 1st week | 4th week | 7th week | 10th week | 13th week | 16th week |
| 0 | T* | 39 | 26 | 59.0 | 57.0 | 61.5 | 61.5 | 60.5 | 57.5 |
| | | 209 | 218 | 45.0 | 43.0 | 44.5 | 43.5 | 35.5 | 34.0 |
| | D* | † — | — | — | — | — | — | — | — |
| | | 128 | 707 | 42.5 | 41.5 | 41.0 | 39.5 | 32.0 | 29.5 |
| | | 376 | 951 | 146.5 | 141.5 | 147.0 | 144.5 | 128.0 | 121.0 |
| 5 | T | 53 | 16 | 58.0 | 59.5 | 61.5 | 62.0 | 61.0 | 61.0 |
| | | 66 | 21 | 50.0 | 50.5 | 52.5 | 51.5 | 51.5 | 52.0 |
| | D | 31 | 35 | 67.0 | 68.5 | 71.5 | 70.5 | 73.5 | 75.5 |
| | | 105 | 138 | 51.0 | 51.5 | 53.0 | 49.5 | 44.5 | 41.0 |
| | | 255 | 210 | 226.0 | 230.0 | 238.5 | 233.5 | 230.5 | 229.5 |
| 10 | T | 172 | 81 | 46.0 | 47.5 | 51.0 | 54.0 | 52.5 | 53.0 |
| | | 283 | 31 | 52.5 | 58.0 | 62.5 | 62.0 | 62.0 | 64.0 |
| | D | 53 | 154 | 48.5 | 49.0 | 53.5 | 52.0 | 49.0 | 47.5 |
| | | 56 | 16 | 68.0 | 68.5 | 72.0 | 67.0 | 67.5 | 65.5 |
| | | 564 | 282 | 215.0 | 223.0 | 239.0 | 235.0 | 231.0 | 230.0 |
| 20 | T | 37 | 13 | 61.0 | 62.5 | 66.0 | 65.0 | 63.0 | 61.0 |
| | | 143 | 23 | 72.5 | 75.0 | 82.0 | 80.0 | 79.0 | 81.0 |
| | D | 127 | 45 | 59.0 | 58.5 | 66.5 | 60.5 | 62.5 | 60.0 |
| | | 107 | 94 | 44.5 | 43.5 | 47.5 | 45.0 | 45.0 | 46.0 |
| | | 414 | 175 | 237.0 | 239.5 | 262.0 | 250.5 | 249.5 | 248.0 |
| 30 | T | 164 | 43 | 46.0 | 47.0 | 51.5 | 51.5 | 46.0 | 45.5 |
| | | † — | — | — | — | — | — | — | — |
| | D | 123 | 7 | 51.5 | 56.0 | 57.0 | 57.5 | 55.5 | 55.5 |
| | | 781 | 58 | 43.0 | 44.5 | 48.5 | 50.5 | 51.5 | 50.0 |
| | | 1,068 | 108 | 140.5 | 147.5 | 157.0 | 159.5 | 153.0 | 151.0 |
| 40 | T | 300 | 336 | 40.0 | 37.5 | 41.0 | 40.5 | 35.0 | 36.0 |
| | | 125 | 34 | 52.0 | 51.5 | 56.0 | 51.5 | 52.5 | 52.5 |
| | D | † — | — | — | — | — | — | — | — |
| | | 62 | 20 | 57.5 | 58.0 | 60.0 | 60.5 | 60.5 | 61.0 |
| | | 487 | 390 | 149.5 | 147.0 | 157.0 | 152.5 | 148.0 | 149.5 |
| 50 | T | † — | — | — | — | — | — | — | — |
| | | 159 | 61 | 52.0 | 56.5 | 62.5 | 62.5 | 61.0 | 62.0 |
| | D | 200 | 10 | 53.0 | 55.5 | 59.5 | 62.5 | 61.0 | 57.5 |
| | | 110 | 8 | 49.0 | 48.0 | 52.0 | 52.5 | 51.5 | 49.5 |
| | | 469 | 79 | 154.0 | 160.0 | 174.0 | 177.5 | 173.5 | 169.0 |

* T = Tablets, D = Drench. † died.

OUTSIDE FLOCKS : INITIAL AND FINAL TOTAL EGG COUNTS, AND LAMB WEIGHTS AT 3-WEEKLY INTERVALS .

(Initial egg counts, sums of three counts on separate days, have been doubled to make them comparable with final egg counts, which are sums of six counts. For statistical computation the tabulated initial counts should therefore be halved.)

WEYBRIDGE *D*

| Dose in Gm. | Administration | TOTAL EGG COUNTS | | WEIGHTS IN LB. AT: | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Initial ×2 | Final ×1 | 1st week | 4th week | 7th week | 10th week | 13th week | 16th week |
| 0 | T* | 140 | 11 | 60.0 | 66.0 | 66.0 | 59.5 | 65.0 | 65.0 |
| | | † — | — | — | — | — | — | — | — |
| | D* | 26 | 12 | 70.0 | 73.0 | 70.0 | 70.5 | 80.0 | 79.0 |
| | | 128 | 14 | 71.0 | 74.5 | 68.5 | 64.5 | 75.0 | 73.0 |
| | | 294 | 37 | 201.0 | 213.5 | 204.5 | 194.5 | 220.0 | 217.0 |
| 5 | T | 134 | 13 | 75.0 | 78.5 | 78.0 | 72.5 | 80.0 | 74.5 |
| | | 298 | 126 | 72.0 | 74.0 | 73.0 | 71.0 | 79.5 | 80.0 |
| | D | 168 | 111 | 59.0 | 60.0 | 61.0 | 59.5 | 65.0 | 67.0 |
| | | 222 | 104 | 64.0 | 67.0 | 68.0 | 64.0 | 71.0 | 71.5 |
| | | 822 | 354 | 270.0 | 279.5 | 280.0 | 267.0 | 295.5 | 293.0 |
| 10 | T | 192 | 23 | 88.0 | 89.0 | 86.5 | 81.5 | 88.0 | 84.5 |
| | | 224 | 29 | 62.0 | 64.5 | 62.0 | 61.0 | 65.0 | 62.0 |
| | D | 338 | 36 | 80.0 | 82.0 | 84.0 | 78.0 | 84.5 | 83.0 |
| | | 228 | 6 | 65.0 | 71.0 | 69.5 | 64.0 | 70.5 | 72.0 |
| | | 982 | 94 | 295.0 | 306.5 | 302.0 | 284.5 | 308.0 | 301.5 |
| 20 | T | 112 | 16 | 59.0 | 64.0 | 64.0 | 60.0 | 69.0 | 65.5 |
| | | 292 | 32 | 72.0 | 75.0 | 71.0 | 71.5 | 81.0 | 77.0 |
| | D | 874 | 13 | 61.0 | 64.0 | 64.5 | 64.5 | 73.0 | 69.0 |
| | | 136 | 33 | 63.0 | 71.0 | 70.0 | 70.0 | 80.0 | 81.0 |
| | | 1,414 | 94 | 255.0 | 274.0 | 269.5 | 266.0 | 303.0 | 292.5 |
| 30 | T | 230 | 13 | 69.0 | 71.0 | 69.0 | 70.5 | 78.0 | 76.5 |
| | | 310 | 17 | 82.0 | 87.0 | 85.5 | 81.0 | 88.0 | 81.5 |
| | D | 118 | 39 | 63.0 | 67.0 | 71.0 | 67.0 | 71.5 | 72.5 |
| | | 296 | 20 | 64.0 | 61.0 | 64.0 | 62.0 | 68.0 | 69.0 |
| | | 954 | 89 | 278.0 | 286.0 | 289.5 | 280.5 | 305.5 | 299.5 |
| 40 | T | 194 | 16 | 74.0 | 80.0 | 79.5 | 73.5 | 81.0 | 75.0 |
| | | 290 | 16 | 63.0 | 69.0 | 69.0 | 68.0 | 75.5 | 73.0 |
| | D | 186 | 8 | 49.0 | 55.0 | 58.0 | 60.0 | 65.0 | 64.0 |
| | | 114 | 42 | 61.0 | 56.5 | 57.5 | 58.0 | 65.0 | 66.5 |
| | | 784 | 82 | 247.0 | 260.5 | 264.0 | 259.5 | 286.5 | 278.5 |
| 50 | T | 96 | 43 | 58.0 | 64.5 | 66.0 | 61.0 | 72.0 | 71.0 |
| | | 422 | 25 | 63.0 | 64.5 | 65.0 | 62.5 | 71.0 | 70.0 |
| | D | 302 | 16 | 60.0 | 61.5 | 65.0 | 62.0 | 71.0 | 70.0 |
| | | 374 | 64 | 71.0 | 76.5 | 77.0 | 71.0 | 83.0 | 83.0 |
| | | 1,194 | 148 | 252.0 | 267.0 | 273.0 | 256.5 | 297.0 | 294.0 |

* T = Tablets, D = Drench. † died.

## APPENDIX V. Variation in Samples of Eggs and Small Worms

The sampling distribution of egg counts in a McMaster slide is known from previously published work to agree closely with the theoretically expected Poisson distribution. This has been confirmed in the present trials by testing all the pre-treatment egg counts from the two flocks at St. Albans. The results, tabulated below, show that means and variances are closely similar, as Poisson theory requires.

TABLE I. VARIANCE OF EGG COUNTS

| FLOCK | EGGS | SUM OF COUNTS | $n$ | MEAN COUNT | SUM OF SQUARES | $n'$ | MEAN SQUARE |
|---|---|---|---|---|---|---|---|
| Inside | N | 47 | 252 | **0.1865** | 29 | 168 | **0.1746** |
| | S | 576 | 252 | **2.2857** | 416 | 168 | **2.4762** |
| | Others | 7,083 | 252 | **28.107** | 4,899 | 168 | **29.161** |
| | Total ... ... | 7,706 | 756 | **10.193** | 5,344 | 504 | **10.604** |
| Outside | All ... ... | 5,409 | 252 | **21.464** | 3,495 | 168 | **20.804** |
| Grand Total ... | ... ... ... | 13,115 | 1,008 | **13.011** | 8,839 | 672 | **13.154** |

The left half of the Table shows the total counts (sums of 9 counts from each of 28 lambs) and mean counts of the eggs of *Nematodirus* (N), *Strongyloides* (S), and " Others", with the total of these three kinds of eggs, for the inside flock, and also the counts for the outside flock (not differentiated). The right half shows the sum of squares of deviations of counts from the respective means of 3 counts per day per lamb ; each group of 3 counts contributes 2 degrees of freedom ($n'$), or 168 for the flock ; the sums of squares divided by the degrees of freedom give the mean squares or variances. In a Poisson distribution the variance is numerically equal to the mean, and the close agreement of each mean in the Table with its variance indicates a satisfactory egg counting technique.

The number of small nematodes present in a given lamb is estimated by sampling the intestinal contents (see Section 2k, p. 12). The technique aims at counting a relatively small number of worms distributed at random in a relatively large volume of water ; hence the same Poisson distribution would be expected *a priori* as with egg counts. Apart from the possibility of insufficient mixing of the contents, the only reason to question this expectation lies in the fact that *Nematodirus* occurs in the small intestine, and this worm has the curious habit of forming dense clusters, often of several hundred worms, and so firmly held together by the worms' heads that shaking will not usually break them up. Under such conditions the variation between parallel counts from the same suspension might well be abnormally high. This has been tested on 50 counts of worms from the small intestine of 17 lambs from the St. Albans inside flock, there being two, three, or four parallel counts from each lamb.

The data are given in the accompanying Table (II). Individual counts, $x$, from a given lamb have been compared with the expected mean count, $\bar{x}$ for that lamb by means of $\chi^2$ where

$$\chi^2 = \frac{S(x-\bar{x})^2}{\bar{x}}$$

The sum of the 17 values of $\chi^2$ is 43.4837, and the total degrees of freedom, i.e. the sum of the degrees of freedom ($n'-1$) for each lamb, is 33. Fisher's Table of $\chi^2$ does not

extend beyond $n = 30$, but for higher values the following expression may be used as a normal deviate with unit variance :—

$$\sqrt{2\chi^2} - \sqrt{2n-1}$$

If this difference is less than 2 the value of the total $\chi^2$ is not abnormally high. In the present case the difference was 1.26. The variances of the counts differ from their respective means by an amount well within the bounds of random variation and these worm counts are therefore distributed, like the egg counts, in reasonable agreement with the Poisson distribution.

TABLE II. VARIANCE OF SMALL-WORM COUNTS : INSIDE FLOCK

| LAMB | COUNTS $x$ 1 | 2 | 3 | 4 | TOTAL $S(x)$ | MEAN $\bar{x}$ | $S(x-\bar{x})^2$ | $\chi^2$ | $n = (n'-1)$ |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 15 | 22 | 27 | 16 | 80 | 20 | 94 | 4.7000 | 3 |
| 2 | 12 | 16 | 10 | 8 | 46 | 11.5 | 35 | 3.0435 | 3 |
| 3 | 47 | 62 | — | — | 109 | 54.5 | 112.5 | 2.0642 | 1 |
| 4 | 45 | 37 | 35 | — | 117 | 39 | 56 | 1.4359 | 2 |
| 5 | 20 | 32 | 24 | 29 | 105 | 26.25 | 84.75 | 3.2286 | 3 |
| 6 | 23 | 22 | 24 | 21 | 90 | 22.5 | 5 | 0.2222 | 3 |
| 7 | 42 | 34 | 35 | — | 111 | 37 | 38 | 1.0270 | 2 |
| 8 | 18 | 14 | 9 | 14 | 55 | 13.75 | 40.75 | 2.9636 | 3 |
| 9 | 63 | 61 | — | — | 124 | 62 | 2 | 0.0323 | 1 |
| 10 | 78 | 87 | — | — | 165 | 82.5 | 40.5 | 0.4909 | 1 |
| 11 | 23 | 35 | 42 | — | 100 | 33.3 | 184.67 | 5.5401 | 2 |
| 12 | 31 | 37 | 53 | — | 121 | 40.3 | 258.67 | 6.4133 | 2 |
| 13 | 63 | 107 | — | — | 170 | 85 | 968 | 11.3882 | 1 |
| 14 | 64 | 60 | — | — | 124 | 62 | 8 | 0.1290 | 1 |
| 15 | 27 | 22 | 24 | 24 | 97 | 24.25 | 12.75 | 0.5258 | 3 |
| 16 | 81 | 77 | — | — | 158 | 79 | 8 | 0.1013 | 1 |
| 17 | 43 | 47 | — | — | 90 | 45 | 8 | 0.1778 | 1 |
| | | | | | | | | 43.4837 | 33 |

$2\chi^2 = 86.9674$ $\quad \sqrt{2\chi^2} = 9.3256$

$2n-1 = 65$ $\quad \sqrt{2n-1} = 8.0623$

Difference $= 1.2633$

## APPENDIX VI

### Dosage Mortality Curve based on Egg Counts at Edinburgh

The egg count comparisons of Table 1 (p. 19) give the ratios of observed to expected counts when the ratio for the control group at each centre is made 100. The expected counts have no regard to level of dosage, which shows its effect in the discrepancy between the tabulated values and 100. In other words, the expectations ignore dosage which, as befits a null hypothesis, is assumed to have no effect, this assumption being disproved by the *deviations* from expectation. Further and more precise comparisons are not possible unless at this stage the real effects of dosage are taken into account. This involves finding a formula which shall provide expectations giving a minimum deviation from the observed values, the expectations being calculable from the dosage.

A very important class of toxicological data can be accurately represented by the " Dosage Mortality " curve. In this the logarithm of the dose is taken to be proportional to the deviate of a normal distribution, the probability of exceeding which gives the

expected frequency of survival of the organism. As applied to the present problem, this means the expected fraction to which the egg-laying activity of the parasites is reduced.

As measured by egg counts, the response at Edinburgh was so regular that it was thought desirable to fit a dosage mortality curve to the data. Standard methods of fitting such a curve to data of this kind, where the experimental animal is appreciably variable but expectations are calculable from prior observations, have not been published. However, the values of the appended Table have been shown to provide nearly as good a fit as is possible with any curve of this type.

In the column of the Table headed " Normal Deviate " the values are logarithmic functions of the dosage (thus, each of the deviates corresponding to the geometric series 5, 10, 20, and 40 gm. differs from its neighbours by a constant amount, 0.24). The next column, " Dosage Factor ", gives the fractional area of the Normal curve lying beyond the deviate shown. The products of the Dosage Factors and the expectations used in compiling Table I give the values headed " Proportional Expectation ", which are proportionately increased in the column " Actual Expectation ", so that their sum is equal to the sum of the " Observed Values ". The final column gives the difference between observed and expected counts.

TABLE I. COMPARISON OF FINAL EGG COUNTS WITH EXPECTATIONS DERIVED FROM A " DOSAGE MORTALITY " CURVE : EDINBURGH

| DOSAGE GM. | NORMAL DEVIATE | DOSAGE FACTOR | PROPORTIONAL EXPECTATION | ACTUAL EXPECTATION | OBSERVED VALUE | DEVIATION |
|---|---|---|---|---|---|---|
| 0 | $-\infty$ | 1.0000 | 105.68 | 620.65 | 620 | − 0.65 |
| 5 | 0.50 | 0.30854 | 50.918 | 299.04 | 335 | +35.96 |
| 10 | 0.74 | 0.22965 | 37.922 | 222.71 | 168 | −54.71 |
| 20 | 0.98 | 0.16354 | 13.176 | 77.38 | 79 | + 1.62 |
| 30 | 1.1204 | 0.13127 | 20.183 | 118.53 | 154 | +35.47 |
| 40 | 1.22 | 0.11123 | 12.698 | 74.57 | 61 | −13.57 |
| 50 | 1.2973 | 0.097266 | 9.045 | 53.12 | 49 | − 4.12 |
| | | | | 1466.00 | 1466 | 0.00 |

A marked feature of the egg counts (Table 1, p. 19) was the very large reduction at the lowest dosage of 5 gm., but the present Table shows that the observed count at this dosage was actually higher than expectation. The principal negative deviation occurs at the 10 gm. dosage. The low values, expected and observed, at 20 gm. are due to the loss of one of the four lambs from this group. The trend of the values as a whole indicates that very high doses would be needed to secure a much larger reduction in the egg count.

From the calculation of regression (of logarithmic final on logarithmic initial count) expected values are available not only for each dosage group but also for each individual lamb. These expectations, also, can be multiplied by the appropriate dosage factor from the above Table and the products proportionately increased to give actual " dosage mortality " expectations for each lamb, for comparison with the observed final egg counts. The importance of being able to do this lies in the possibility of detecting minor or subsidiary factors on which the experiment is capable of throwing light.

An examination of the deviations of individual lambs in the Edinburgh flock enabled a comparison to be made between those given tablets and those drenched, as shown in Table II. It will be seen that, at all dosage levels except 10 gm., the final egg count was lower than expected for tablets and higher for drenches. Excluding the untreated controls, the total negative deviation for 12 lambs given tablets was about 91 eggs, and of course

the drenched lambs show a corresponding positive deviation (exact numerical equality between these totals can be achieved only by including the controls). This suggests that a given weight of drug in the form of tablets is equivalent to a somewhat higher weight given as a drench. This effect was not confirmed at other centres and should probably be ascribed to chance.

The importance of calculating expectations for individual lambs lies not only in the possibility of detecting such minor factors, but also in circumventing difficulties arising from the loss of experimental animals. Thus, owing to the death of a lamb in the 20 gm. group, the observed egg count is the sum of only three lamb-counts ; no problem arises since the total expectation for that group is similarly the sum of expectations for the three survivors.

TABLE II. COMPARISON OF FINAL EGG COUNTS WITH EXPECTED FOR INDIVIDUAL LAMBS : TABLETS *versus* DRENCHES

| DOSAGE GM. | TABLETS | | | DRENCHES | | |
|---|---|---|---|---|---|---|
| | EXPECTED | OBSERVED | DEVIATION | EXPECTED | OBSERVED | DEVIATION |
| 5 | 60.94 | 38 | −22.94 | 59.88 | 66 | 6.42 |
| | 131.61 | 136 | 4.39 | 46.91 | 95 | 48.09 |
| | | | −18.55 | | | 54.51 |
| 10 | 49.00 | 47 | − 2.00 | 74.57 | 41 | −33.57 |
| | 51.83 | 43 | − 8.83 | 47.31 | 37 | −10.31 |
| | | | −10.83 | | | −43.88 |
| 20 | 21.88 | 21 | − 0.88 | 30.02 | 41 | 10.98 |
| | 25.48 | 17 | − 8.48 | (*Lamb* | *died*) | — |
| | | | − 9.36 | | | 10.98 |
| 30 | 15.91 | 21 | 5.09 | 39.22 | 55 | 15.78 |
| | 46.19 | 31 | −15.19 | 17.21 | 47 | 29.79 |
| | | | −10.10 | | | 45.57 |
| 40 | 18.81 | 2 | −16.81 | 22.85 | 39 | 16.15 |
| | 20.73 | 18 | − 2.73 | 12.18 | 2 | −10.18 |
| | | | −19.54 | | | 5.97 |
| 50 | 18.78 | 4 | −14.78 | 10.65 | 37 | 26.35 |
| | 12.75 | 5 | − 7.75 | 10.94 | 3 | − 7.94 |
| | | | −22.53 | | | 18.41 |
| | Total Effect ... | | −90.91 | | | 91.56 |

## APPENDIX VII

### Full Analysis of Weight Data : Outside Flocks

Since in previous data from St. Albans, the initial egg count had proved extremely valuable in increasing the precision of comparisons of final live weight, this variable was from the first used with the six outside flocks of the co-operative experiment. In these experiments the response to phenothiazine was striking and apparently uniform from

flock to flock. The question naturally arises as to whether it is, so far as can be detected, uniform for different degrees of initial infestation.

This question is of some importance for various reasons :—

(*a*) Since the drug acts primarily by diminishing infestation it might be expected to confer a greater benefit, measured in live weight, on heavily than on lightly infested sheep. In this case, if the flocks chosen for these experiments were, as is to be expected, somewhat more heavily infested than average lambs of the same age, the advantage to be obtained by more widespread dosing would be somewhat less than the advantage shown in these experiments.

(*b*) The disadvantage of a heavy infestation, as measured in final weight, would be different for different levels of dosage. The process of making the same allowance for initial egg count at all dosage levels would, therefore, contribute less to the precision of the final comparisons, than if allowance could have been made in a more accurate form.

(*c*) Some previous workers had reported that the response to phenothiazine, measured in live weight, was actually less with the more heavily infested sheep. As the drug appears to be without effect on the growth of uninfested sheep, this implies that with infestation increasing from zero, the effect of the drug increases to a maximum and then diminishes, and that the degree of infestation in the experiments reported exceeded the degree showing maximal response.

In order to clear up, so far as the data allow, doubts of this kind, each of the six flocks was re-analyzed, using simultaneous regression on four variates, namely :—

(1) initial weight ($w$) ;
(2) initial egg count ($e$) ;
(3) product of dosage and initial egg count ($1{,}000x$) ;
(4) dosage ($d$).

### Computational procedure for determining the Regression Coefficients for four independent variates

We shall first set out the procedure for a single flock, choosing for the purpose the Outside Flock at Edinburgh, which contained 31 sheep.

We require to express the final weight $y$ as accurately as possible in terms of the four variables $w$, $e$, $x$ and $d$, which are used as independent variates. The expected weight Y will thus be expressed in the form

$$Y = c + b_1 w + b_2 e + b_3 x + b_4 d.$$

#### Computation

The sum of squares and products for the four independent variates form the *Information matrix*, as shown below :—

| | $w$ | $e$ | $x$ | $d$ |
|---|---|---|---|---|
| $w$ | 1429.0968 | —2161.2257 | —44.891288 | —178.22581 |
| $e$ | | 119266.19380 | 513.60468 | —9400.8065 |
| $x$ | | | 97.002534 | 767.05968 |
| $d$ | | | | 9874.1935 |

The sums of products for $y$, the dependent variate, are :—

| | |
|---|---|
| S $(wy)$ | 1073.5157 |
| S $(ey)$ | —3664.8708 |
| S $(xy)$ | 52.496454 |
| S $(dy)$ | 851.12903 |

The sum of squares for $y$ is 1577.4194.

The $4 \times 4$ matrix is next inverted by treating its terms as the coefficients of four equations with four unknowns corresponding with the four independent variates used, the right-hand side of these equations being first 1, 0, 0, 0. (*See* Statistical Methods, Sec. 29.)

The stages in inversion are :—

| | | | | | |
|---|---|---|---|---|---|
| (i) | 14079.414 | −23015.827 | −306.55543 | = | 9.8741935 |
| | −23015.827 | 1089282.319 | 12282.412 | = | 0 |
| | −306.55543 | 12282.412 | 369.44125 | = | 0 |
| (ii) | 5107.5400 | −4737.7559 | | = | 3.6479344 |
| | −4737.7559 | 251568.18 | | = | 0 |
| (iii) | 1262.4482 | | | = | 0.91770422 |

The four solutions provide the elements of the first row and column of the inverted matrix, known as the covariance matrix :—

$.0^{3}72692425$ ($c_{11}$) $.0^{4}1369008$ ($c_{12}$) $.0^{3}14804893$ ($c_{13}$) $.0^{4}14653560$ ($c_{14}$)
$.0^{4}1369008$
$.0^{3}14804893$
$.0^{4}14653560$

Substituting these values we next solve the three further sets of equations, using the same coefficients, but the three sets :—

| | | |
|---|---|---|
| 0 | 0 | 0 |
| 1 | 0 | 0 |
| 0 | 1 | 0 |
| 0 | 0 | 1 |

as the right-hand sides.

The complete inverted matrix is then :—

| | | | |
|---|---|---|---|
| $.0^{3}72692425$ ($c_{11}$) | $.0^{4}1369008$ ($c_{12}$) | $.0^{3}14804893$ ($c_{13}$) | $.0^{4}14653560$ ($c_{14}$) |
| $.0^{4}1369008$ ($c_{21}$) | $.0^{4}1475860$ ($c_{22}$) | $-.0^{3}4793034$ ($c_{23}$) | $.0^{4}5153320$ ($c_{24}$) |
| $.0^{3}14804893$ ($c_{31}$) | $-.0^{3}4793034$ ($c_{32}$) | .04278510 ($c_{33}$) | $-.0^{2}3777339$ ($c_{34}$) |
| $.0^{4}14653560$ ($c_{41}$) | $.0^{4}5153200$ ($c_{42}$) | $-.0^{2}3777339$ ($c_{43}$) | $.0^{3}4440361$ ($c_{44}$) |

The regression coefficients are calculated from this matrix by multiplying the four columns by the sums of products with the dependent variate, e.g. $(wy) = 1073.5157$, and adding for each column ; thus giving :—

$$b_w = .7504363\,;\quad b = -.02069319\,;\quad b_x = .946581\,;\quad b_d = .0065078$$

The sum of squares accounted for by the regression for 4 degrees of freedom is found by multiplying the regression coefficients by the same sums of products, and adding. In this case the analysis is therefore :—

| | *d.f.* | *s.s.* | *Mean square* |
|---|---|---|---|
| Regression ... ... | 4 | 936.674 | 234.168 |
| Remainder, error ... | 26 | 640.745 | 24.644 |
| | 30 | 1577.419 | |

The sampling variance for any Regression coefficient is found by multiplying the mean square error by the corresponding term in the covariance matrix. For example, the variance of $b_x$ is $24.644 \times c_{33}$, which equals 1.0544. Hence the standard error of $b_x$ is $\pm$ 1.027.

The average improvement in weight due to the drug is

$$\left( b_x \times \frac{\bar{e}}{1000} \right) + b_d$$

which is 0.10385 lb. per gm. of drug.

---

To examine the homogeneity of the six flocks in the response they have shown, the same formula was fitted also to the pooled data from all six.

The sums of squares of deviations within flocks, taking all flocks together, is 13,615.035 for 154 degrees of freedom, corresponding to 1577.4194 for 30 degrees of freedom at Edinburgh. The regressions for the pooled results were :—

$$b_w = .9108725\ ;\ b_e = -.0^2657879\ ;\ b_x = .1491275\ ;\ b_d = .09088997$$

The improvement in weight due to the drug, estimated from the pooled results, is therefore 0.10831 lb. per gm. of drug compared with 0.10385 lb. per gm. at Edinburgh.

The sum of squares accounted for by fitting the fourfold regression formula is 9,923.516 for 4 degrees of freedom, corresponding to 936.674 for 4 degrees of freedom at Edinburgh, leaving 3691.519 for 150 d.f. in the residue.

| | *d.f.* | *s.s.* |
|---|---|---|
| Regression ... | 4 | 9923.516 |
| Remainder ... | 150 | 3691.519 |
| | 154 | 13615.035 |

However, when regressions are fitted separately to the six flocks the residual errors for 130 d.f. amount to only 2898.143, hence the 20 d.f. for heterogeneity in regression contain 793.376 of the sum of squares.

Putting these facts together we have :—

| | *d.f.* | *s.s.* | *Mean square* |
|---|---|---|---|
| Average regression ... | 4 | 9923.516 | |
| Heterogeneity in regressions | 20 | 793.376 | 39.669 |
| Residual error ... ... | 130 | 2898.143 | 22.293 |
| Totals ... ... | 154 | 13615.035 | |

It is clear from the contrast in the mean squares that the different flocks are largely heterogeneous, at least in some aspects of the appropriate prediction formulae. Since it appears from a cursory examination that there are real differences in the regressions on initial weight, and since such differences are to be expected with flocks differing somewhat in their feeding and management, the next test was designed to ascertain whether the whole of the heterogeneity shown by the different flocks is due to their regression on initial weight, so that their response to the other three variates involving initial egg count and dosage is homogeneous.

So far the method of analysis has been of a standard form of which the theory and computational details are available in such a text book as Fisher's *Statistical Methods, Section* 29. The method of procedure for fitting regressions to a series of samples which shall be pooled in respect of some variates, though fitted independently in respect to others, has not been published, at least in so accessible a form.

We first remove the variate $w$, regression on which is not to be pooled. This is done by replacing any sum of squares, such as $S(e^2)$ by the reduced value :

$$S\ (e^2)\ -\ \frac{S^2\ (ew)}{S\ (w^2)}$$

The sum of products, including products with $y$, are reduced in the same way ; e.g. we replace S($ey$) by

$$S(ey) - \frac{S(ew)\ S(wy)}{S(w^2)}$$

With the flock from Edinburgh, for example, we have the reduced values :—

| | *e* | *x* | *d* | *y* |
|---|---|---|---|---|
| *e* | 115997.77 | 445.71550 | —9670.3377 | —2041.3905 |
| *x* | | 95.592393 | 761.46119 | 86.218105 |
| *d* | | | 9851.9666 | 985.00954 |
| *y* | | | | 771.01088 |

This reduction is carried out for each of the six flocks, and the corresponding terms from each added to give an Information Matrix of the pooled results :—

| | *e* | *x* | *d* | *y* |
|---|---|---|---|---|
| *e* | 1135493.4 | 25626.228 | 1593.5787 | 1094.737 |
| *x* | | 1432.7898 | 5159.2389 | 623.19722 |
| *d* | | | 46165.529 | 4821.2357 |
| *y* | | | | 3743.5904 |

This 3 × 3 matrix is then inverted by the same method as shown for the 4 × 4 matrix for Edinburgh, and the covariance matrix of the pooled results is thus obtained.

| | | |
|---|---|---|
| $.0^5 2637967$ ($c_{11}$) | $-.0^4 7840451$ ($c_{12}$) | $.0^5 8671054$ ($c_{13}$) |
| $-.0^4 7840451$ ($c_{21}$) | $.0^3 3498232$ ($c_{22}$) | $-.0^3 3882393$ ($c_{23}$) |
| $.0^5 8671054$ ($c_{31}$) | $-.0^3 3882393$ ($c_{32}$) | $.0^4 6474964$ ($c_{33}$) |

From this the regression coefficients are obtained as already explained, giving :—

$$b_e = .004168397\,; \qquad b_x = .22246295\,; \qquad b_d = .079716148$$

The improvement in weight due to the drug calculated on these results is 0.10570 lb. per gm. The sum of squares accounted for by fitting a general regression on the 3 variates is 518.405 for 3 degrees of freedom, which when subtracted from the average regression for 4 degrees of freedom from the pooled results gives the general regression on initial weight as 9405.111 for 1 degree of freedom.

| | *d.f.* | *s.s.* | *Mean square* |
|---|---|---|---|
| General regression on 4 variates ... ... ... | 4 | 9923.516 | |
| General regression on 3 variates ... ... ... | 3 | 518.405 | |
| Additional contribution due to *w*. ... ... | 1 | 9405.111 | |
| Heterogeneity of regression on initial weight ... | 5 | 466.434 | 93.287 |
| Total due to *w* in 6 flocks together ... ... | 6 | 9871.545 | |

This analysis clears up the whole situation, as shown by the table below, in which the effects of initial weight are separated from those of the other three variates :—

| | *d.f.* | *s.s.* | *Mean square* |
|---|---|---|---|
| Error... ... ... ... ... ... ... | 130 | 2898.143 | 22.293 |
| Heterogeneity of regression in *e.x.d.* ... ... | 15 | 326.942 | 21.796 |
| General regression in *e.x.d.* ... ... ... | 3 | 518.405 | 172.802 |
| Additional contribution due to general regression on *w*. ... ... ... ... ... ... | 1 | 9405.111 | |
| Heterogeneity of regression on *w*. ... ... | 5 | 466.434 | 93.287 |
| | 154 | 13615.035 | |

It is obvious that there is no heterogeneity in the response to the variates other than initial weight, but that there is strong heterogeneity in this one respect, and a significant general response to the other three variates used simultaneously.

These three degrees of freedom may now be further sub-divided into one portion representing the response to dosage when initial egg count is ignored, while the remaining two degrees of freedom will show any further predictive value which the initial egg count may have had in these experiments, when used in conjunction with dosage. The first of these is calculated from the pooled information matrix, using the regression of final weight on drug (ignoring initial egg count), namely :—

$$\frac{4821.2357}{46165.529} = .10443 \text{ lb. per gm.},$$

giving the contribution to the sum of squares :—

$$\frac{(4821.2357)^2}{46165.529} = 503.499,$$

so that the subdivision is numerically :—

| | *d.f* | *s.s.* |
|---|---|---|
| Dosage only ... ... ... ... ... ... ... | 1 | 503.499 |
| Additional portion accounted for by using initial egg count ... | 2 | 14.906 |
| | 3 | 518.405 |

It is therefore concluded that variations in initial egg count have in these data added nothing to the precision when initial weight is properly taken into account, and that the reduced increase in weight to be expected from heavily infested lambs is quite adequately anticipated by taking account of the reduced weight with which the lambs enter the experiment. We obtain further important confirmation of the uniformity of the response to dosage in the six different flocks used. The estimate of the average response to drug given in Section 4c (p. 21) of the REPORT is also fully confirmed by the more comprehensive analysis of this Appendix.

## Legends to Figures 1–12 (pages 49 to 53)

Dosage and logarithmic Worm Count.
Solid line—Tablets ; Broken line—Drenches.
Pooled data from 4 Inside Flocks.

Fig. 1—*Haemonchus* (Class A)
Fig. 2—*Trichostrongylus axei* (Class B)
Fig. 3—*Ostertagia* (Class B)
Fig. 4—*Bunostomum* (Class C)
Fig. 5—*Cooperia* (Class C)
Fig. 6—*Trichostrongylus* spp. (Class C)
Fig. 7—*Chabertia* (Class D)
Fig. 8—*Oesophagostomum* (Class D)
Fig. 9—*Strongyloides* (Class E)
Fig. 10—*Trichuris* (Class E)
Fig. 11—*Nematodirus* (Class F)
Fig. 12—*Moniezia* (Class F)

## Legend to Figure 13 (page 54)

Time and Weight-gains for each rate of dosing with Phenothiazine.
Pooled data from 6 Outside Flocks.

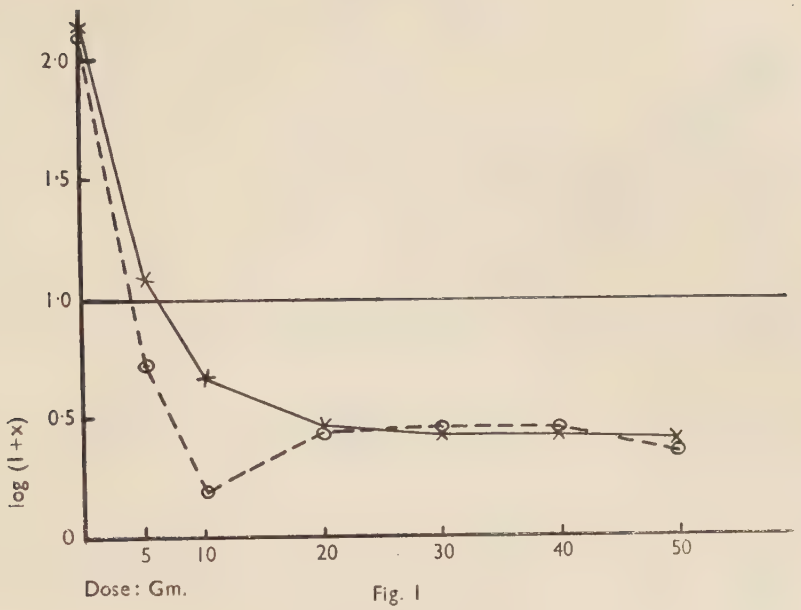


Fig. 1

*Haemonchus* (Class A)

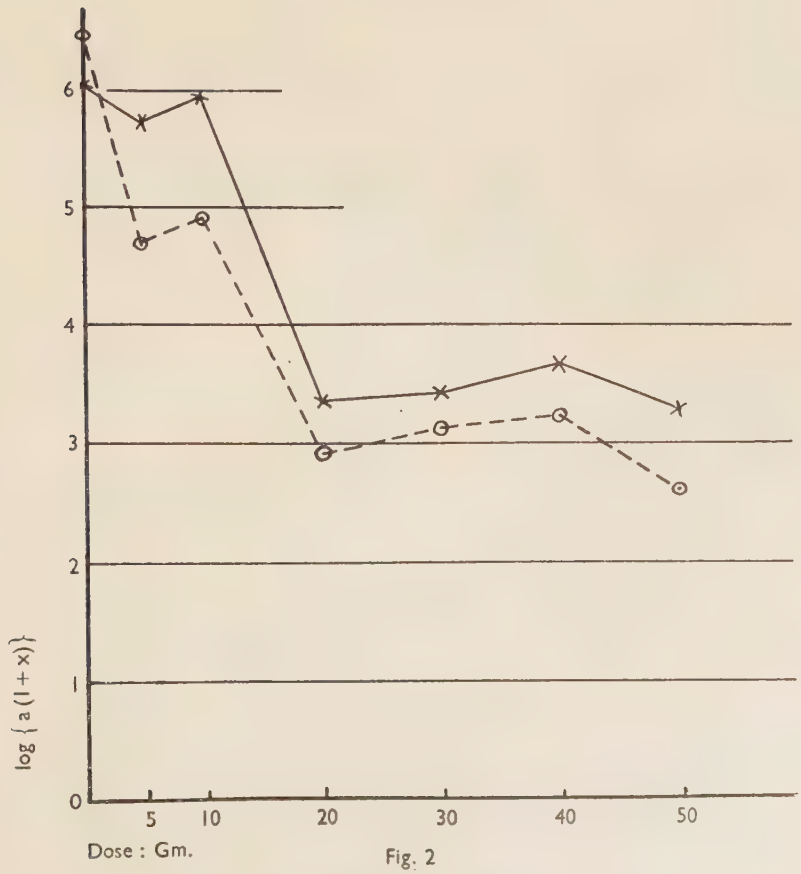


Fig. 2

*Trichostrongylus axei* (Class B)

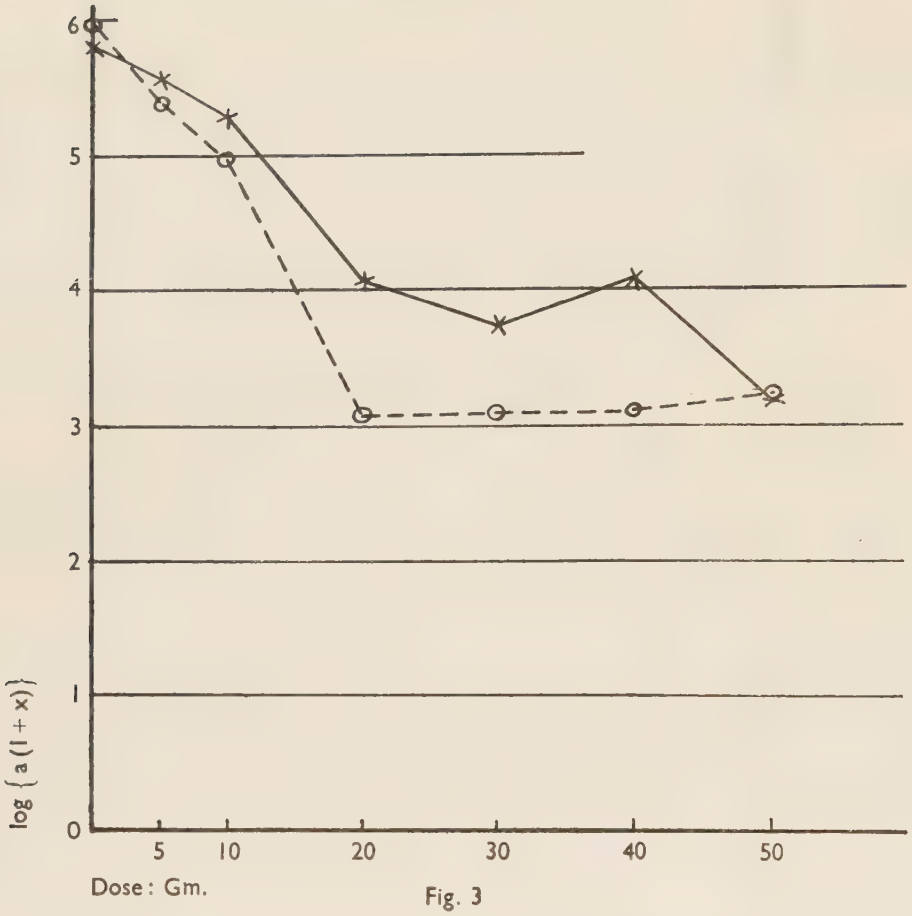


Fig. 3

*Ostertagia* (Class B)

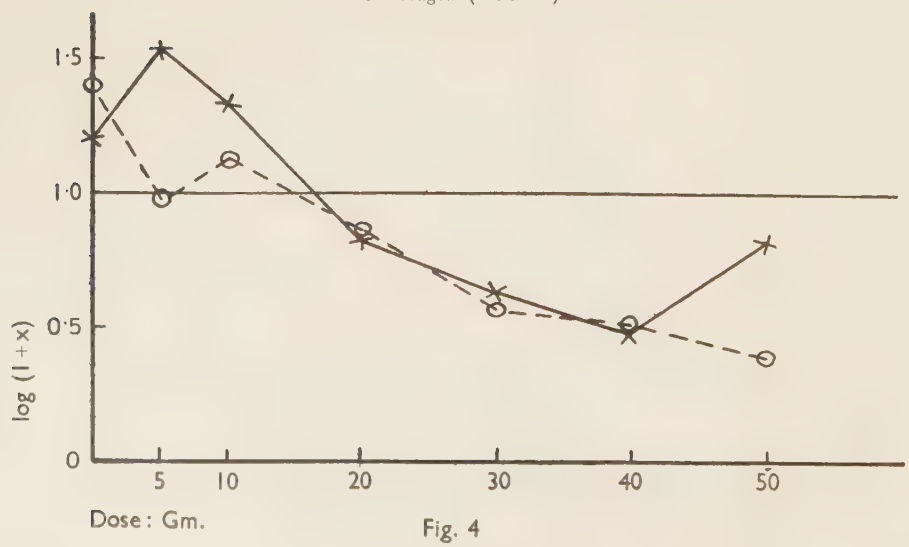


Fig. 4

*Bunostomum* (Class C)

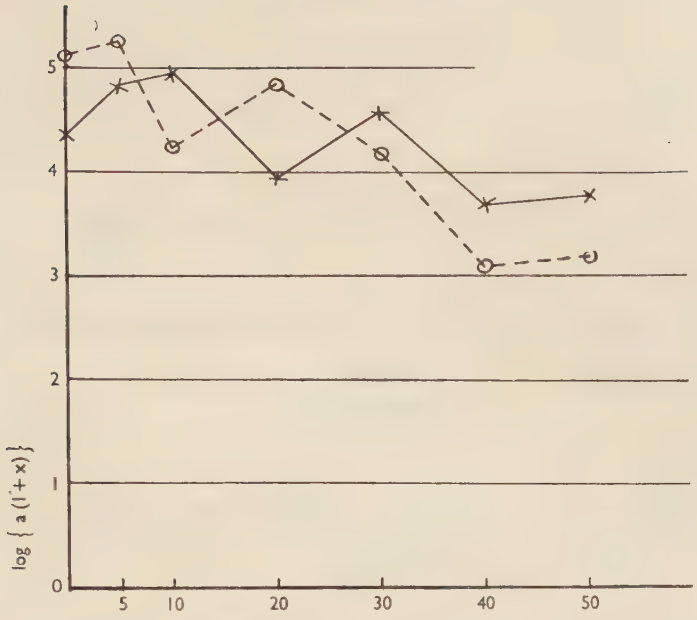


Fig. 5
*Cooperia* (Class C)

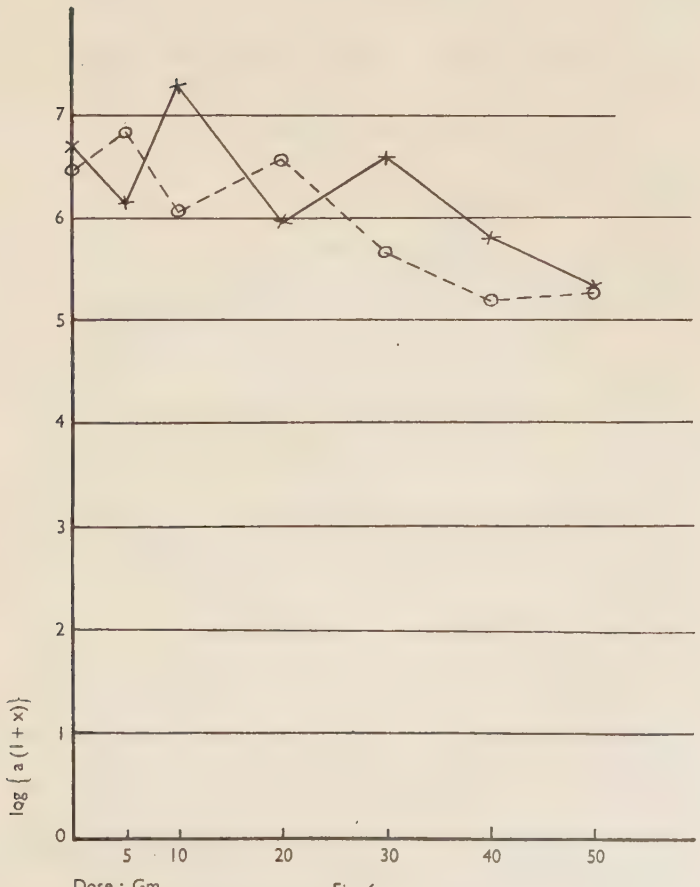


Fig. 6
*Trichostrongylus* spp. (Class C)

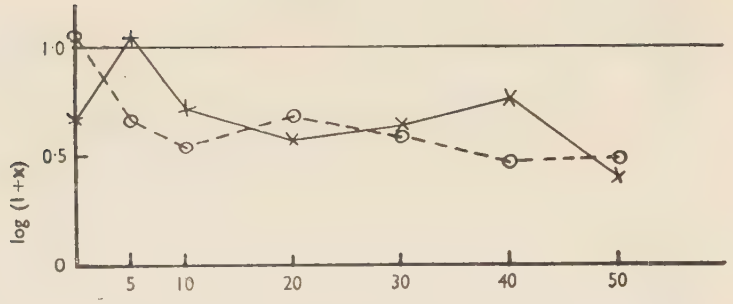


Fig. 7
*Chabertia* (Class D)

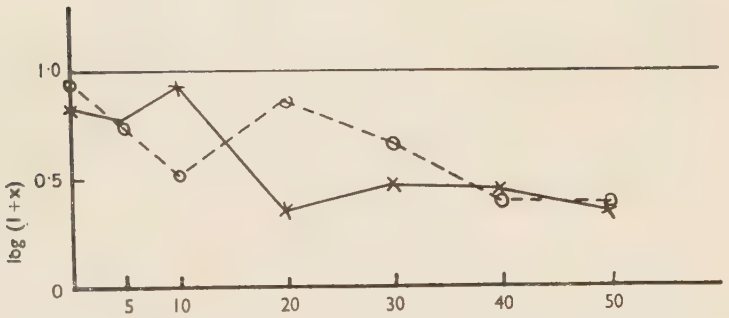


Fig. 8
*Oesophagostomum* (Class D)

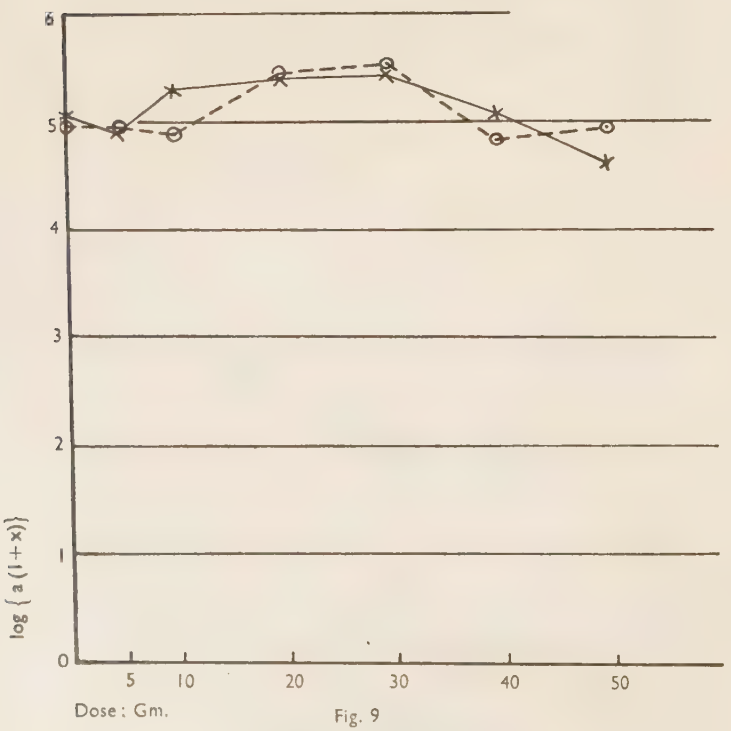


Fig. 9
*Strongyloides* (Class E)

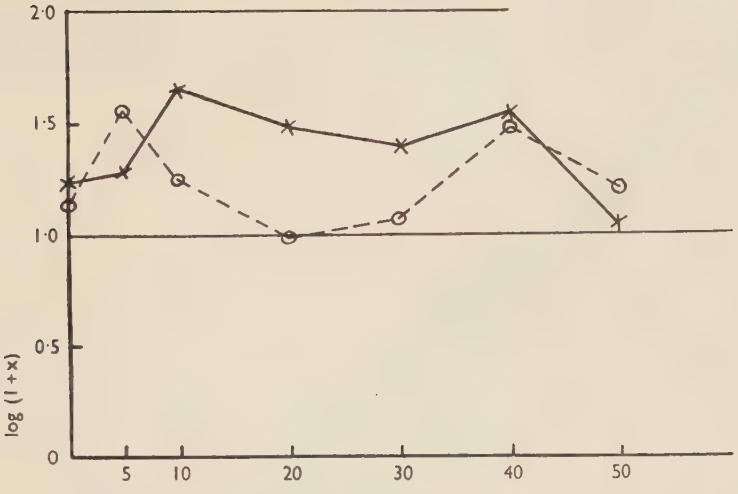


Fig. 10
*Trichuris* (Class E)

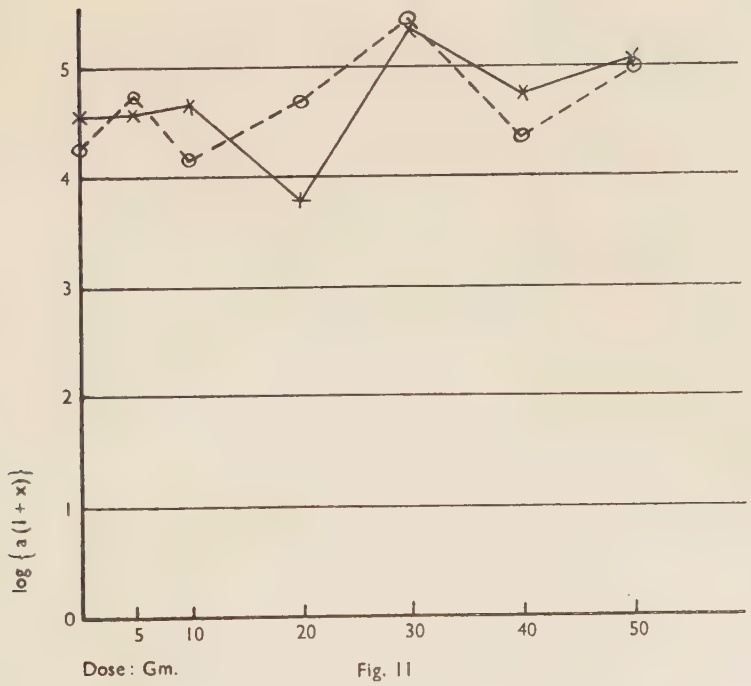


Fig. 11
*Nematodirus* (Class F)

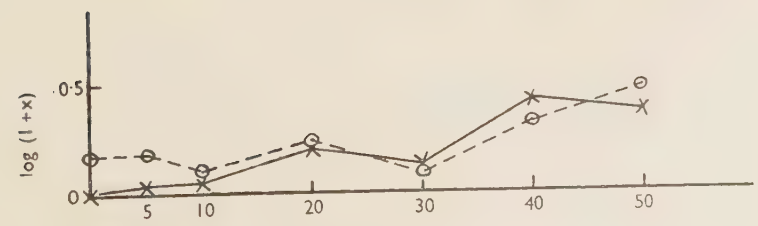


Fig. 12
*Moniezia* (Class F)

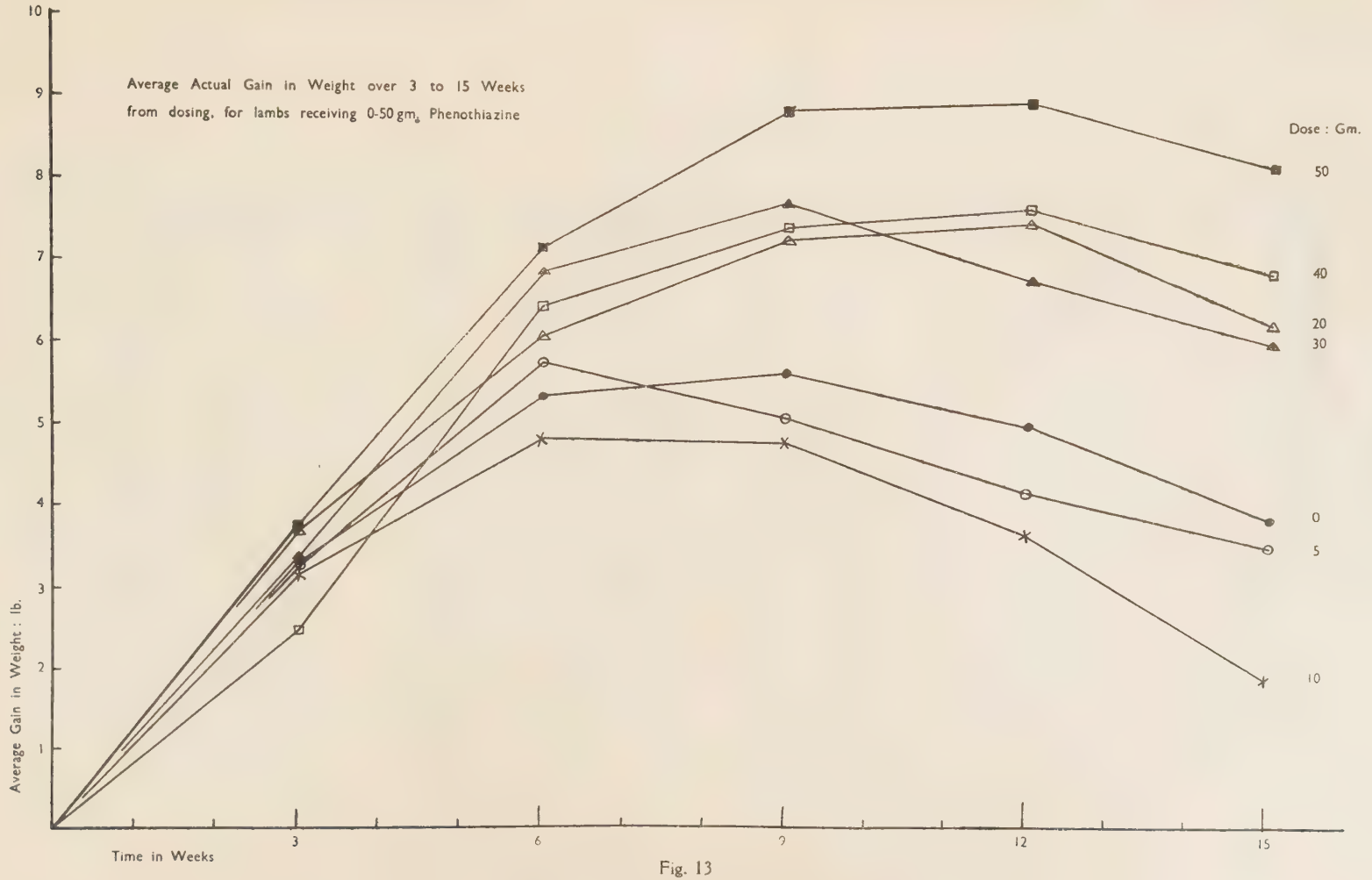


Fig. 13

*Time and Weight-gains for each rate of dosing with Phenothiazine*
*Pooled data from 6 Outside Flocks*

## REFERENCES


1. BODDIE, G. F., CORNER, H. H., MORGAN, D. O. & SLOAN, J. E. N., 1941.—" Field trials with phenothiazine on lambs." *Vec. Rec.*, **53** (12), 170–173.

2. COLLIER, H. B., 1940.—" Enzyme inhibition by derivatives of phenothiazine and of sulphanilamide." *Canad. J. Res., Sect. B*, **18** (11), 345–350.

3. DAVEY, D. G. & INNES, J. R. M., 1942.—" The present position of phenothiazine as an anthelmintic." *Vet. Bull., Weybridge*, **12** (8), R7–R14.

4. ERRINGTON, B. J., 1941.—" Phenothiazine as an equine anthelmintic." *Vet. Med.*, **36** (4), 188–193.

5. FISHER, R. A., 1941.—*Statistical methods for research workers*. Edinburgh & London, 8th Edit., xv + 344 pp.

6. GORDON, H. McL., 1939.—" The anthelmintic efficiency of phenothiazine." *Aust. vet. J.*, **15** (6), 245–252.

7. GORDON, H. McL., 1939a.—" A preliminary note on the chemotherapy of oesophagostomiasis of sheep with special reference to the efficiency of phenothiazine." *J. Coun. sci. industr. Res. Aust.*, **12** (3), 203–206. Erratum **12** (4), 347.

8. GORDON, H. McL., 1939b.—" A preliminary note on the anthelmintic efficiency of phenothiazine against *Trichostrongylus* spp. in sheep." *J. Coun. sci. industr. Res. Aust.*, **12** (4), 345–347.

9. GORDON, H. McL., 1940.—" The anthelmintic efficiency of phenothiazine against immature *Haemonchus contortus*." *J. Coun. sci. industr. Res. Aust.*, **13** (4), 245–246.

10. GORDON, H. McL., & WHITTEN, L. K., 1939.—" A preliminary note on the anthelmintic efficiency of phenothiazine against *Haemonchus contortus* in sheep." *J. Coun. sci. industr. Res. Aust.*, **12** (3), 207.

11. GORDON, H. McL. & WHITTEN, L. K., 1940.—" A field trial comparing phenothiazine, tetrachlorethylene emulsion, and copper sulphate and nicotine sulphate mixture for the treatment of trichostrongylosis." *J. Coun. sci. industr. Res. Aust.*, **13** (2), 81–85.

12. GRAHAME, T., MORGAN, D. O., & SLOAN, J. E. N., 1940.—" Report on phenothiazine as an anthelmintic in horses." *Vet. Rec.*, **52** (36), 660–663.

13. HABERMANN, R. T. & HARWOOD, P. D., 1939.—" Anthelmintic efficacy of unconditioned crude phenothiazine." [Abstract of paper presented at the 15th Annual Meeting of the American Society of Parasitologists.] *J. Parasit.*, **25** (6), Suppl. p. 36.

14. HABERMANN, R. T., HARWOOD, P. D. & HUNT, W. H., 1941.—" Critical tests with phenothiazine as an anthelmintic in horses." *N. Amer. Vet.*, **22** (2), 85–92.

15. HABERMANN, R. T. & SHORB, D. A., 1942.—" The effect of phenothiazine-salt mixtures on the development of parasite larvae in the feces of sheep." *N. Amer. Vet.*, **23** (5), 318–321.

16. HARWOOD, P. D., HABERMANN, R. T. & JERSTAD, A. C., 1939.—" Efficacy of commercial phenothiazine in the removal of roundworms from sheep." *Vet. Med.*, **34** (7), 440–443.

17. HARWOOD, P. D., HABERMANN, R. T. & SWANSON, L. E., 1939.—" Phenothiazine as an anthelmintic." [Abstract of paper presented at the 15th Annual Meeting of the American Society of Parasitologists.] *J. Parasit.*, **25** (6), Suppl. p. 34.

18. HARWOOD, P. D., JERSTAD, A. C. & SWANSON, L. E., 1938.—" The efficacy of phenothiazine for the removal of ascarids and nodular worms from swine." [Abstract of paper presented at the 14th Annual Meeting of the American Society of Parasitologists.] *J. Parasit.*, **24** (6), Suppl. pp. 16–17.

19. HATCHER, W. L., 1941.—" Phenothiazine poisoning in horses." *N. Amer. Vet.*, **22** (3), 159–160.

20. HOLMAN, H. H. & PATTISON, I. H., 1941.—" A study of the blood picture of lambs suffering from parasitic gastritis." *Vet. Rec.*, **53** (34), 491–498.

21. IMPERIAL BUREAU OF AGRICULTURAL PARASITOLOGY (HELMINTHOLOGY), 1942.—" Bibliography of phenothiazine as an anthelmintic." St. Albans, 6 pp.

22. KNIPLING, E. F., 1938.—" Internal treatment of animals with phenothiazine to prevent development of hornfly larvae in the manure." *J. econ. Ent.*, **31**, 315–316.

23. KNOWLES, R. H. & FRANKLIN, A. V., 1940.—" Treatment of equine strongylosis by phenothiazine." *Vet. Rec.*, **52** (36), 663–664.

24. LAPAGE, G., 1940.—" Experiments on the anthelmintic action of phenothiazine." *Vet. Rec.*, **52** (36), 648–657.

25. LEIPER, J. W. G. & PETERS, B. G., 1941.—" The effect of phenothiazine on the weights of worm-free sheep and goats." *J. Helminth.*, **19** (3/4), 71–74.

26. LEIPER, J. W. G., PETERS, B. G. & CLAPHAM, P. A., 1941.—" A controlled experiment with phenothiazine in sheep." *J. Helminth.*, **19** (1/2), 25–34.

27. McCULLOCH, E. C., 1942.—" Phenothiazine-medicated pellets for range sheep." *J. Amer. vet. med. Ass.*, **101** (785), 114–119.

28. McEWEN, A. D., 1940.—" Comparative tests on the treatment of lambs with phenothiazine and with copper sulphate and nicotine sulphate." *Vet. Rec.*, **52** (36), 657–658.

29. MÖNNIG, H. O., 1942.—" Warning against the worm-remedy ' phenothiazine '." *Fmg S. Afr.*, **17** (197), 487–488.

30. MONTGOMERIE, R. F., 1940.—" Trial of phenothiazine in ascarid infestation in dogs." *Vet. Rec.*, **52** (36), 665.

31. PETERS, B. G. & LEIPER, J. W. G., 1941.—" Single *versus* multiple doses of phenothiazine in lambs." *J. Helminth.*, **19** (3/4), 63–70.

32. PETERS, B. G., LEIPER, J. W. G. & CLAPHAM, P. A., 1941.—" A phenothiazine experiment statistically treated." *J. Helminth.*, **19** (1/2), 9–24.

33. ROBERTS, F. H. S., 1939.—" The value of phenothiazine in the treatment of oesophagostomiasis in sheep." *Aust. vet. J.*, **15** (6), 237–244.

34. ROBERTS, F. H. S., 1939a.—" A preliminary note on the treatment of oesophagostomiasis in sheep with phenothiazine." *J. Coun. sci. industr. Res. Aust.*, **12** (3), 208.

35. ROBERTS, F. H. S., 1941.—" Phenothiazine as an anthelmintic for horses, cattle and pigs." *Aust. vet. J.*, **17** (4), 130–137.

36. ROWLANDS, W. T., 1940.—" Phenothiazine in parasitic gastritis." *Vet. Rec.*, **52** (36), 658–660.

37. ROWLANDS, W. T. & HARBOUR, H. E., 1941.—" ' Pining ' in lambs on permanent pasture in N. Wales. An investigation into the cause of the condition." *Vet. Rec.*, **53** (11), 153–156.

38. SCHMIDT, H., CHRISTIAN, T. T. & SMOTHERMAN, W. M., 1941.—" Is phenothiazine poisonous to horses? " *J. Amer. vet. med. Ass.*, **99** (774), 225–229.

39. SHAW, J. N., 1941.—" Use of phenothiazine in Oregon sheep." *N. Amer. Vet.*, **22** (5), 280–283 .

40. SHORB, D. A., HABERMANN, R. T. & HEEMSTRA, L. C., 1941.—" Comparison of the efficacy of phenothiazine and copper sulphate-nicotine sulphate mixture as anthelmintics for sheep." *Vet. Med.*, **36** (10), 502–506.

41. STEWART, W. L. & CROFTON, H. D., 1941.—" Parasitic gastritis in sheep. Comparative trials on lambs with phenothiazine and copper-nicotine mixture." *Vet. Rec.*, **53** (12), 167–170.

42. SWALES, W. E., 1939.—" Tests of phenothiazine, a highly efficient anthelmintic. On a means of administration and the indicated uses for the control of parasitic diseases of sheep." *Canad. J. comp. Med.*, **3** (7), 188–194.

43. SWALES, W. E., SYLVESTRE, P. E. & WILLIAMS, S. B., 1942.—" Field trials of control measures for parasitic diseases of sheep." *Canad. J. Res., Sect. D*, **20** (5), 115–132.

44. TAYLOR, E. L. & SANDERSON, K. M., 1940.—" Phenothiazine—a remarkably efficient anthelmintic." *Vet. Rec.*, **52** (36), 635–647.

45. WOLFE, W. R. & DENNIS, W. R., 1941.—" Toxemia resulting from the use of phenothiazine." *Vet. Bull. U.S. Army*, **35** (3), 171–174.

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